Phytomedicine ▮ (▮▮▮▮) ▮▮▮–▮▮▮

Contents lists available at ScienceDirect

Phytomedicine

journal homepage: www.elsevier.de/phymed

Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies ☆

M. Wasif Saif [a,*], F. Lansigan [a], S. Ruta [a], L. Lamb [a], M. Mezes [a], K. Elligers [a], N. Grant [a], Z.-L. Jiang [a], S.H. Liu [b], Y.-C. Cheng [a]

[a] Yale University School of Medicine, Section of Medical Oncology, 333 Cedar Street, FMP 116 New Haven, CT 06520, USA
[b] PhytoCeutica Inc., New Haven, CT, USA

ARTICLE INFO

Keywords:
Capecitabine
PHY906
Herbal medicines
Diarrhea

ABSTRACT

Purpose: The botanical formulation, PHY906, has been used widely in Eastern countries to treat gastrointestinal symptoms including diarrhea, nausea and vomiting. PHY906 may also have anti-tumor properties and may potentiate the action of several chemotherapeutic agents based on pre-clinical studies. We conducted a Phase I study using PHY906 in combination with capecitabine in patients with advanced pancreatic and gastrointestinal malignancies to determine the maximum tolerated dose (MTD) of capecitabine in combination with PHY906.

Patients and Methods: This study was a single institution, open-label, Phase I study of PHY906 800 mg BID on days 1-4 in combination with escalating doses of capecitabine (1000, 1250, 1500, and 1750 mg/m^2) orally twice daily on days 1-7 of a 14-day cycle (7/7 schedule). Capecitabine was increased until the appearance of dose limiting toxicities (DLTs). Measurements of efficacy included tumor response by Response Evaluation Criteria in Solid Tumors (RECIST).

Results: Twenty-four patients with a median age of 67 years (range 40-84) with pancreatic cancer (15), colon cancer (6), cholangiocarcinoma (1), esophageal cancer (1) and unknown primary (1) received a total of 116 cycles (median 5 cycles; range 1-17 cycles) over 4 dose levels of capecitabine. One DLT (Grade 4 AST/ALT, Grade 3 hyponatremia) was observed in the 1000 mg/m^2 cohort of patients. No further DLT was observed in the subsequent cohorts and doses of capecitabine were escalated to 1750 mg/m^2 BID. There were no DLTs at the maximum dose level of 1750 mg/m^2, however, the delivered dose-intensity of capecitabine was similar at the 1750 mg/m^2 dose level as the 1500 mg/m^2 dose level. Therefore, the MTD was defined at 1500 mg/m^2 of capecitabine in this dosing schedule with PHY906. One patient achieved a partial response, and 13 patients had stable disease that lasted more than six weeks.

Conclusion: The MTD of capecitabine was determined to be 1500 mg/m^2 BID administered in a 7/7 schedule, in combination with PHY906 800 mg BID on days 1-4. This combination was well tolerated and warrants further study.

Introduction

Pancreatic cancer is the fourth most common malignancy in the United States and most newly diagnosed individuals will die within a year (Jemal et al., 2007). Gemcitabine is the current standard of care for both the adjuvant treatment of pancreatic cancer and the treatment of advanced stage disease (APC). In the adjuvant setting, gemcitabine confers an estimated 5-year disease-free survival of only 16%; therefore, even with surgically resectable pancreatic cancer, most patients will relapse after 5 years (Neuhaus et al., 2008). In APC, patients treated with gemcitabine have a median survival of approximately 6 months (Burris et al., 1997).

Second-line chemotherapy after gemcitabine is needed for patients who maintain a good performance status and can tolerate further treatment. 5-FU-based chemotherapy was determined to have activity in APC well before the gemcitabine-era. Response rates to 5-FU are usually less than 5% responses are, partial, and survival is less than 6 months. Nevertheless, 5-FU still has a role in the palliation of gemcitabine-refractory pancreatic cancer (Milella et al., 2004; Mitry et al., 2006; Tsavaris et al., 2005).

☆ Presented as a Poster at the American Society of Clinical Oncology Gastrointestinal Cancers Symposium, 2008.

* Corresponding author. Tel.: +1 203 737 1875; fax: +1 203 785 3788.
E-mail address: wasif.saif@yale.edu (M.W. Saif).

0944-7113/$ - see front matter © 2010 Published by Elsevier GmbH.
doi:10.1016/j.phymed.2009.12.016

Capecitabine (Xeloda) is an orally administered flouropyrimidine that is converted to 5-FU in tumor tissue and mimics the action of continuous intravenous infusion of 5-FU. Capecitabine is currently approved by the US Food and Drug Administration (FDA) for treating, as first-line single-agent therapy, patients with advanced or metastatic colorectal cancer, when single-agent fluoropyrimidine therapy is preferred. It is also approved for use in metastatic breast cancer patients as either a single agent following resistance to both anthracycline-based and paclitaxel-based regimens, in those where further anthracycline treatment is contraindicated, or in combination with docetaxel after the failure of earlier anthracycline-based chemotherapy. Most recently, it was approved for use in Dukes C colon cancer patients who have complete surgical resection of the tumor, as single-agent adjuvant therapy, when treatment with fluoropyrimidine alone is preferred. A Phase II study by Boeck et al., demonstrated that capecitabine 1250 mg/m^2 twice daily for 14 days every 3 weeks in gemcitabine-pretreated patients with advanced pancreatic cancer was effective in disease stabilization (39%), and resulted in a median time to progression of 2.3 months and median overall survival of 7.6 months (Boeck et al., 2007). There were no objective responses in this study, however. The convenience of oral administration has added advantages as a palliative chemotherapeutic option, however, the known toxicities of capecitabine such as diarrhea and hand-foot syndrome (HFS), may compromise its efficacy. In the USA, it is difficult to administer capecitabine 1250 mg/m^2 twice daily for 14 days without experiencing toxicities requiring dose reduction.

PHY906 is a botanical formulation composed of four distinct herbs: *Scutellaria baicalensis Georgi, Glycyrrhiza uralensis Fisch., Ziziphus jujuba Mill., and Paeonia lactiflora Pall.* (Table 1). This herbal formula has been documented in Asia to treat a variety of ailments such as abdominal cramps, fever, headache, vomiting, thirst, and diarrhea for over 1700 years. Anti-diarrheal activity was demonstrated in a previous clinical study of PHY906 with irinotecan and 5-FU/leucovorin in colorectal cancer (CRC). PHY906 was well tolerated up to 2.4 g/day and the frequency of diarrhea and vomiting was significantly lower with PHY906 than with placebo treatment (Farrell and Kummar 2003). Additionally, PHY906 has been demonstrated to have antitumor activity in a pancreatic cancer mouse model. PHY906 is active in potentiating the activity of capecitabine in nude mice bearing PANC-1 tumors (Saif et al., 2007) .

Table 1
Herbal Ingredients in the Dried Aqueous Extract of the Traditional Chinese Medicine Formulation PHY906.

Scientific Name	Common Name	Traditional Use
Scutellaria baicalensis Georgi.	Scute Baical Skullcap Root	Used to reduce capillary permeability; reduce inflammation; treat enteritis and dysentery; increase the secretion of bile to treat jaundice; relieve muscle spasms to treat coughing; expel parasites.
Glycyrrhiza uralensis Fisch.	Licorice Root	Used to moisten the lungs and stop coughs; relax spasm and stop pain; moderate the action of herbs; reduce heartburn and release toxins.
Ziziphus jujuba Mill.	Date Fruit	Has diuretic and strengthening effects.
Paeonia lactiflora Pall.	White Peony Root	Used to suppress and soothe pain: soothe ligaments, and detoxify the blood.

CHARACTERIZATION OF PHY906 FOR QUALITY CONTROL (QC) PURPOSES

PHY906 is comprised of a traditional hot water extract of four commonly used herbs, *Scutelleria baicalensis Georgi* (S), *Paeonia lactiflora Pall.* (P), *Glycyrrhiza uralensis Fisch.* (G), and *Ziziphus jujuba Mill.* (Z), in the ratio of 3:2:2:2, respectively. This extract consists of a complex mixture of multiple phytochemicals with multiple biological and pharmacological properties. At this time it is not possible to identify the subset of relevant biologically active phytochemicals from the complete mixture. For this reason we utilize high level chemical and biological metrics to characterize the PHY906 product.

The raw ingredients of PHY906 are pre-selected to meet rigid specifications set by PhytoCeutica for acceptance by the herbal manufacturer, Sun Ten Pharmaceuticals in Taiwan. The PHY906 extract is comprised of greater than 75% low molecular weight phytochemical compounds less than 1000 amu, 10% macromolecular components including protein, nucleic acid and complex carbohydrates and 5% water. In addition, 10% by weight of excipient insoluble cellulose is added during the spray dry step in manufacturing. Heavy metals (Pb, Hg, Cd, As) are all less than 0.5 ppm, with mercury and cadmium less than 0.03 ppm, as detected by atomic absorption measurements. Pesticides levels (BHCs, DDTs, PCNB) are less than 0.2 ppm by LC-MS or GC-MS. Total bacteria counts are 260 cfu/g while E. coli and Salmonella species are not detected. Over 90%, by weight of PHY906, excluding water content (5%) and insoluble starch excipient (10%), can be re-extracted. The final PHY906 liquid extract (100 mg/ml) is stable for 18 hours at room temperature and the properly stored bulk dry extract (vacuum packed, light tight and 4 °C) appears to be stable for more than three years.

Chemical fingerprint analysis is conducted by capillary LC coupled Mass Spectrometry using a reverse phase C-18 column and a QTOF-II MS instrument (specific conditions given in figure legend 1) The fingerprint separates in the first dimension by chemical property, in the second dimension by exact mass and in the third dimension the intensity of each ion with mass between 50-1500 amu. Thirty-nine individual phytochemical peaks with intensities greater than 1% of the largest peak are used to define the overall chemical fingerprint pattern. These thirty-nine phytochemical peaks make up more than 85% of the total ion current and 25 of the compounds are identified by the use of marker compounds, exact mass comparison and ms/ms fragmentation. Most of the molecular ions in the fingerprint arise from S (63%) followed by G (26%), P (8%) and Z (3%). The thirty-nine peaks are uniquely identified to one of the four herbs and hence form a signature of both the composition, extraction method and the herbal ratios. In addition to the chemical fingerprint, up to four marker compounds for each herbal ingredient are used for absolute quantitation; baicalin (S), baicalein (S), scutellarin (S), wogonin (S), glycyrrhizin (G), liquirtin (G), paeoniflorin (P), albiflorin (P). Using commercial marker standards for six of these compounds, we can use absolute quantitation to determine the milligrams of phytochemical per gram of dry PHY906 powdered extract. The combination of chemical fingerprint that provides a global view of the phytochemicals and absolute quantitation of a subset of phytochemical markers provides a comprehensive chemical analysis of PHY906. Spectral data are shown in Fig. 1 tabulated peaks used in the chemical fingerprint are given in Table 2 (submitted to Journal of Chinese Medicine).

The biological fingerprint analysis monitors the differential gene expression profile of a HepG2 cell line that is exposed to a standardized one IC50 concentration (0.85 mg/ml) of PHY906 extract for a period of 24 hours. Using the Affymetric U133A chip, approximately 18,000 genes are monitored. From three

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016

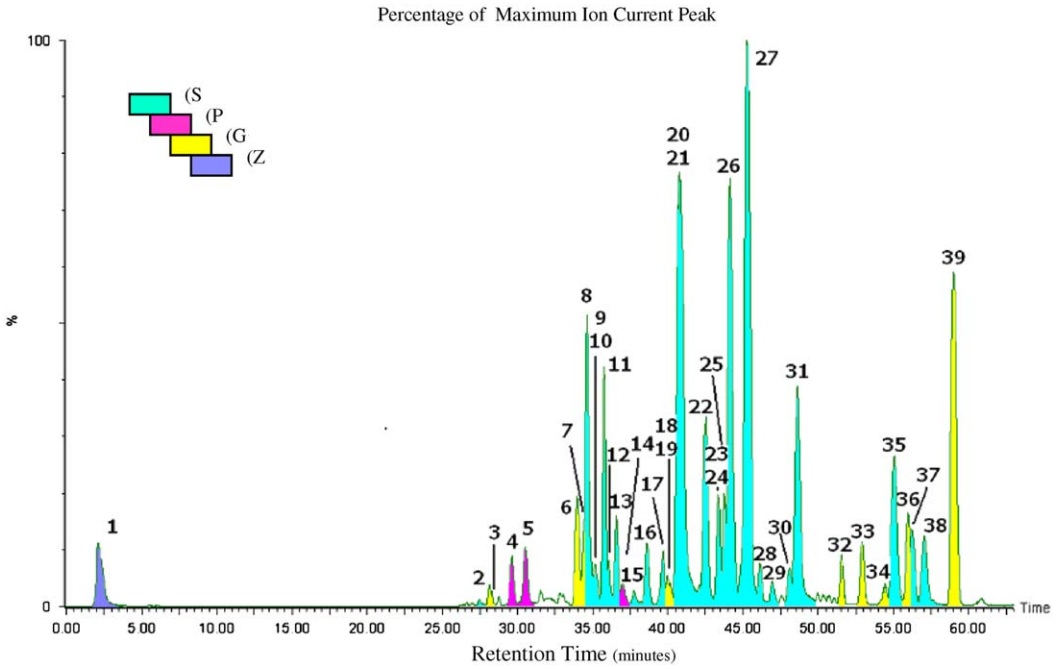

Fig. 1. LC/MS(+) spectrum of PHY906 extract and herbal source identification. Thirty-six peaks are resolved and 64 compounds have been identified or tentatively identified (21). Thirty-nine compound peaks are used to define the chemical fingerprint for batch-to-batch comparisons. Of the 39 peaks of the chemical fingerprint of PHY906 (S) accounts for 25 of 39 peaks, (P) accounts for 3 of 39 peaks, (G) accounts for 10 of 39 peaks and (Z) accounts for only 1 of 39 peaks. All of the peaks identified are associated with a single herbal ingredient. The single identified peak for (Z) is very hydrophilic, has no UV chromophore, elutes in the solvent front of the C18 reverse phase column and ionizes only in (+) positive MS mode. The total ion count for the spectrum is 2.9e4. The complete chemical fingerprint of 39 peaks account for more than 82% of the total ions above a threshold of 1.5% of the largest peak.

Table 2
Chemical Fingerprint Pattern and Identification.

ID	Retention Time	Mass	Herb	Confirmed Chemical Identification	mg of compound/gram dry weight PHY906
1	2.3	159.1	Z		
2	26.57	304	S		
3	26.57	432.1	G		
4	28.93	480.2	P	Albiflorin	
5	30.02	178.1	P	Paeoniflorin	16.3 +/− 0.4
6	33.5	256.1	G	Liquiritin	
7	34.11	256.1	G		
8	34.27	548.1	S		
9	34.9	548.1	S		
10	35.17	324.1	S		
11	35.56	548.1	S		
12	35.85	462.1	S	Scutellarin	1.3 +/− 0.3
13	36.44	548.1	S		
14	36.96	480.2	P		
15	37.76	460.1	S	Wogonoside isomer	
16	38.75	346.1	S		
17	39.86	476.1	S		
18	40.14	430.1	G	Ononin	
19	40.36	256.1	G	Liquiritigenin	
20	41.02	446.1	S	Baicalin	46.4 +/− 2.2
21	41.02	462.1	S		
22	42.91	446.1	S		
23	43.82	446.1	S		
24	43.82	476.1	S		
25	44.25	430.1	S		
26	44.63	460.1	S		
27	45.81	460.1	S		
28	46.67	490.1	S		
29	47.58	270	S	Apigenin	
30	48.76	300.1	S		
31	49.28	270	S	Baicalein	14.0 +/− 1.0
32	52.12	984.5	G	Licorice Saponin A3	
33	53.57	880.4	G		
34	55.22	820.4	G		
35	56.1	284.1	S	Wogonin	7.1 +/− 0.2
36	56.65	838.4	G	Licorice Saponin G2	
37	57.41	374.1	S		
38	58.12	284.1	S		
39	59.67	822.4	G	Glycyrrhizin	14.2 +/− 0.5

Table 3
Gene Response Biofingerprint.

Protein Name	Gene Name	Fold Change from Control
Abhydrolase domain containing 4	ABHD4	2.9
Aldo-keto reductase family 1, member C1	AKR1C1	16.6
Aldehyde dehydrogenase 1 family, member A1	ALDH1A1	2.3
Betaine-homocysteine methyltransferase 2	BHMT2	3.1
CD24	CD24	−2.2
Carnitine palmitoyltransferase 1A	CPT1A	3.1
Cytochrome P450, family 1, subfamily A, polypeptide 1	CYP1A1	275.0
Epithelial membrane protein 2	EMP2	1.7
Fibrinogen alpha chain	FGA2	−1.5
Glutamate-cysteine ligase, modifier subunit	GCLM	7.0
Glutathione peroxidase 1	GPX1	−1.4
Hepcidin antimicrobial peptide	HAMP	2.3
Insulin-like growth factor binding protein 3	IGFBP-3-2	4.3
Keratin 23 (histone deacetylase inducible)	KRT23	−1.9
Oxidative stress induced growth inhibitor 1	OKL38	5.7
Nuclear protein 1	P8	3.0
Phosphoenolpyruvate carboxykinase 1 (soluble)	PCK1	2.6
Pim-1 oncogene	PIM1	2.3
Son of sevenless homolog 1	SOS1	9.0
Tubulin, alpha 1a	TUBA3	−1.6

independent analyses, only 77 genes indicated more than a two fold change between control treatment and PHY906 treated cells. From this set of candidate genes, 20 genes are chosen based on reproducibility, robustness, sensitivity and finally confirmation by qRT-PCR and are given in Table 3. While one should not infer any mechanism of action data from this set of genes, the gene expression pattern, provides a comprehensive fingerprint of PHY906 that clearly complements the chemical analysis fingerprint.

Conventional methods included in the certificate of analysis of PHY906 include sensory evaluation, TLC analysis, total ash, acid-insoluble ash, heavy metal testing, microbial testing and pesticide residues. These tests combined with single marker compound analysis and an extensive chemical fingerprint; provide a robust phytochemical analysis of PHY906. While the chemical fingerprint is comprehensive, the bioresponse gene expression provides a secondary, orthogonal biological method for characterizing PHY906. This bioresponse fingerprint employing HepG2 as the sensor cell; indicate a relatively small number of possible genes, 77 out of 18,000, that are responsive to a high level dose. Of these 77 genes, the gene expression profile of 20 genes provides a robust and unique fingerprint to characterize PHY906 that can be used for defining product reproducibility.

The principle objectives of this study were to determine the maximum tolerated dose (MTD) of PHY906 and capecitabine in patients with APC and other gastrointestinal (GI) malignancies who failed to respond to standard therapy, and to determine the dose-limiting toxicities (DLTs) and non-DLTs of this combination. We also observed the objective response and clinical benefit of this combination.

Patients and Methods

Patient Selection

Entry criteria included patients with histologic or cytologic diagnosis of locally advanced or metastatic pancreatic adenocarcinoma and other GI cancers, who had failed prior chemotherapy. All patients must have had at least one previously unirradiated unidimensionally measureable lesion by computed tomography (CT) or magnetic resonance imaging (MRI) scan of ≥ 20 mm (by conventional CT) or ≥ 10 mm (by spiral CT). Other eligibility criteria included the following: age 18 years or older, Eastern Cooperative Oncology Group (ECOG) performance status of 2 or lower, and adequate bone marrow (absolute neutrophil count [ANC] $\geq 1500/\mu l$, and platelets $>100{,}000/\mu l$), kidney (creatinine ≤ 1.5 mg/dl), and liver function (bilirubin ≤ 2 mg/dl, transaminase levels ≤ 2.5 times the upper limit normal (ULN), alkaline phosphatase ≤ 2.5 times ULN or 5 times ULN for patients with liver metastasis). Prior 5-FU or capecitabine was permitted in this Phase I study. Patients were excluded if they had brain or meningeal metastasis, were taking herbal medicines or supplements ≤ 7 days prior to study entry, had known hypersensitivity or intolerance to either PHY906 or capecitabine, or had any preexisting medical condition of sufficient severity to prevent full compliance with the study. Patients were required to provide written informed consent.

Table 4
Dose escalation scheme.

Dose Level	Dose of Capecitabine day 1-7 q 14 days	Dose of PHY906 x day 1-4 q 14 days	No. of patients	No. of cycles	DLT
1	1000 mg/m^2 BID	800 mg BID	6	30	1
2	1250 mg/m^2 BID	800 mg BID	3	15	0
3	1500 mg/m^2 BID	800 mg BID	7	35	0
4	1750 mg/m^2 BID	800 mg BID	8	36	0

The protocol and associated Informed Consent Forms were reviewed by the Investigational Review Boards (IRB) associated with the study site, and approved prior to study initiation. This study was conducted in accordance with Good Clinical Practice guidelines of the International Conference on Harmonization, and the Declaration of Helsinki.

Treatment plan

The starting dose of capecitabine was 1000 mg/m^2 administered twice daily for 7 days of a 14-day cycle (7/7 dosing schedule). The dose of capecitabine was increased by 250 mg/m^2 in successive cohorts of patients to a planned maximum of 1750 mg/m^2. PHY906 was given at a fixed dose of 800 mg administered twice daily days 1-4 of a 14-day cycle. Responding patients could continue to receive treatment until the appearance of progressive disease or the development of serious toxicity. Patients who developed progressive disease at any time were taken off the study.

At least three patients were enrolled onto each dose level (Table 4). If a dose-limiting toxicity (DLT) was observed after the

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016

first cycle in one patient, three additional patients were accrued onto that dose level. If two or more patients presented a DLT on any dose level, that dose level was considered intolerable, the dose level immediately below was declared the maximum-tolerated dose, and dose escalation ceased. Dose-limiting toxicity was defined using the National Cancer Institute's Common Toxicity Criteria (National Cancer Institute 2006) scale as one or more of the following effects attributable to study drug: Grade 3 or 4 neutropenia complicated by fever, Grade 4 neutropenia lasting longer than 5 days, Grade 4 thrombocytopenia, any other Grade 3-4 non-hematologic toxicity except alopecia, and delay of recovery from treatment-related toxicity for more than 2 weeks.

Dose reduction and omission criteria for both agents were defined for hematologic and non-hematologic toxicity. Doses of capecitabine were omitted if the ANC was $<500/\mu l$ or platelets were $<25{,}000/\mu l$, and then reduced by 250 mg/m^2 if the ANC recovered to $\geq 1{,}000/\mu l$ or if the platelet count was $\geq 50{,}000/\mu l$. There was no replacement of missed doses of capecitabine. Whenever capecitabine was omitted, PHY906 was also held during that period. Treatment was also omitted if any non-hematologic toxicity of Grade 2 or higher emerged and had not resolved to baseline or $\leq$ Grade 1 by scheduled start of treatment. Oral prophylactic antiemetics were administered 30 min prior to each dose of capecitabine. Pyridoxine and emollients were routinely prescribed for all patients who entered the study.

The dose-intensity of capecitabine during the course of treatment in evaluable patients was calculated in mg/m^2 per week by dividing the total amount of capecitabine delivered during the course of treatment by BSA, and by the number of weeks that elapsed between the first dose and last dose.

Pretreatment and follow-up

Before entry onto the study, all patients underwent a full history and physical examination. Complete blood count (CBC) with differential, electrolyte levels, and creatinine levels were measured. Routine chemistry tests, urinalysis, electrocardiograms, CT scans of the chest, abdomen, and pelvis, were performed at baseline in all patients. Additional imaging investigations were performed if clinically indicated or for disease measurement.

While on study, patients were followed for symptoms of toxicity. Patients underwent clinical examinations during which weight, ECOG performance status, and quality of life (QOL) were assessed at the start of each new cycle and at the end of treatment. Complete blood counts with differential, serum chemistry, creatinine level, and electrolyte level, as well as tumor markers were measured at the beginning of each cycle and at study completion.

CT scans and imaging of measurable disease to assess tumor response were performed every three cycles. At completion of the study, all clinical, laboratory, radiological imaging, and other evaluations were repeated. After removal from the study, patients underwent follow-up examinations every 3 months until death. Additional treatment after disease progression was left to the discretion of the treating physician.

Assessment of Efficacy

All patients were included in efficacy measurements on an intent-to-treat basis. Tumor responses were evaluated according to the RECIST Criteria (Therasse et al., 2000). Measurements were made at baseline and after every three cycles of therapy. A complete response (CR) was defined as the disappearance of all target lesions; a partial response (PR) was defined as at least a 30% decrease in the sum of the longest diameter of target lesions; progressive disease (PD) was defined as at least 20% increase in the sum of the longest diameter, or the appearance of one or more new lesions, and/or unequivocal progression of non-target lesions; and stable disease (SD) was defined as neither sufficient shrinkage or sufficient increase to qualify as PR or PD. Any response had to be confirmed no less than 4 weeks later.

Time to tumor progression (TTP) was defined as the date of first treatment to first evidence of radiological disease progression. Duration of overall response was measured from the time of CR or PR until the recurrence or PD. Duration of stable disease was measured from the start of treatment until the criteria for PD were met taking as reference the smallest measurements recorded since treatment started. Best overall response was recorded at the end of the study.

In addition, the Edmonton Symptom Assessment system was used to assess QOL measures (Davison et al., 2006; Moro et al., 2006; Vignaroli et al., 2006). A set of 9 linear 1-10 scales (pain, fatigue, nausea, depression, anxiety, drowsiness, appetite, shortness of breath, and sensation of well-being, and a tenth scale for any symptom of importance to the patient) was recorded prior to each cycle.

Results

Twenty-four patients, whose baseline characteristics are summarized in Table 5, received a total of 116 cycles (median 5 cycles; range 1-17 cycles) over four dose levels of capecitabine. The median age was 67 years (range 40-84). Fifteen patients had APC, six had CRC, one had bile duct cancer, one had esophageal cancer, and one had cancer of unknown primary. All had metastatic cancer. All had received prior regimens (median 2, range 1-5). The mean body surface area (BSA) was 1.87 mg/m^2 (range 1.57-2.41). All subjects were evaluable for safety, and 23 subjects were evaluable for therapeutic benefit. One DLT (Grade 4 AST/ALT, Grade 3 hyponatremia) was observed in the 1000 mg/m^2 cohort of patients. This patient was taken off study, and was included in the response assessment and survival calculations in an intent-to-treat basis. No further DLT was observed in the subsequent cohorts and doses of capecitabine were escalated to 1750 mg/m^2 BID. Two patients at the 1750 mg/m^2 dose level necessitated one dose reduction for diarrhea, and one of these patients also required an additional dose reduction for hand-foot syndrome (HFS). Based on the dose intensity of capecitabine administered at the 1750 mg/m^2 dose level, the 1500 mg/m^2 dose level was expanded for additional safety. The MTD of capecitabine was determined to be 1500 mg/m^2 BID for 7 days of a 14-day cycle, in combination with PHY906 800 mg BID on days 1-4.

Toxicity

Hematologic toxicity was uncommon with no Grade 3-4 toxicities observed. Two patients experienced Grade 1-2 neutropenia (dose levels 3 and 4), and 8 patients experienced Grade 1-2 thrombocytopenia. There were no dose reductions due to hematologic toxicity (Table 6).

Non-hematologic toxicities were mild to severe in nature. The most relevant toxicities per dose level are listed in Table 7. At the 1750 mg/m^2 dose level, one patient experienced Grade 4 diarrhea, and another patient experienced Grade 3 diarrhea, both requiring dose reduction. There were another thirteen patients who reported mild diarrhea symptoms, one of which was Grade 2. Grades 2 and 3 HFS was observed in nine patients (38%, one at

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016

Table 5
Patients' demographic characteristics.

Baseline Characteristics	PHY906 800 mg BID and				Total n = 24
	Capecitabine 1000 mg/m^2 BID n = 6	Capecitabine 1250 mg/m^2 BID n = 3	Capecitabine 1500 mg/m^2 BID n = 7	Capecitabine 1750 mg/m^2 BID n = 8	
Gender					
Male	4	3	7	5	19
Female	2			3	5
Ethnicity					
White	6	3	6	7	22
Black				1	1
Hispanic			1		1
Age (yr)					
Median	67	61	70	61	67
Range	50-81	58-73	50-79	40-84	40-84
Cancer Type					
Pancreatic	4	1	4	6	15
Colon		2	2	2	6
Bile Duct			1		1
Esophogeal	1				1
Unknown primary	1				1
Previous Treatment					
Median prior therapies	2	2	2	1	2
Range	2-3	1-5	1-4	1-4	1-5
ECOG					
0 or 1	6	2	7	7	22
2		1		1	2
BSA (m2)					
Mean	1.80	2.05	1.83	1.91	1.88
Range	1.63-2.05	1.89-2.16	1.70-2.04	1.57-2.41	1.57-2.41
Starting dose capecitabine					
Median dose (mg)	1800	2650	2725	3150	2725

Table 6
Hematologic Toxicities.

Dose level (mg/m^2)	#Patients (cycles)	No. of Patients (Cycles) with Neutropenia		No of Patients (Cycles) with Thrombocytopenia	
		Grade 1-2	Grade 3-4	Grade 1-2	Grade 3-4
1000	6 (30)	0	0	1 (1)	0
1250	3 (15)	0	0	1 (1)	0
1500	7 (35)	1 (2)	0	2 (2)	0
1750	8 (36)	1 (1)	0	4 (4)	0

dose level 1, four at dose level 3, and four at dose level 4), with four patients requiring dose reduction. There were no other dose reductions required for toxicities other than diarrhea and HFS. Sixteen patients experienced anorexia, nine of which were Grade 2. Fifteen patients experienced constipation, three of which were Grade 2. Nine of fifteen patients with constipation did not experience diarrhea during their treatment course; the remainder also reported diarrhea sometime during treatment. Thirteen patients complained of mild nausea and vomiting, and one patient at the 1500 mg/m^2 dose level experienced more severe Grade 3 nausea and emesis thought to be due to non-adherence to prophylactic antiemetics. Nine patients complained of mild xerostomia. Five patients experienced stomatitis, one of which was Grade 3 and resolved after treatment was held for one week. There were five infections reported: one Grade 1 and one Grade 2 fever, one toe infection, one sinus infection, and one Grade 3 oral candidiasis with mucositis that required hospitalization. There were four patients who experienced elevation of transaminases, one of which was Grade 4 and was considered the only DLT of the study.

Efficacy

Twenty-two patients were assessable for response. There was one confirmed PR that lasted 36 weeks in a patient (treated at the 1000 mg/m^2 dose level) with a metastatic cancer of unknown primary, which was most consistent with cholangiocarcinoma based on immunohistochemistry. This responder had received two prior therapies: gemcitabine, and carboplatin/paclitaxel. This patient also had a biochemical response evidenced by a $>65\%$ decrease in all tumor markers (CEA, CA19-9, AFP) during the treatment period. An additional thirteen patients had SD greater than six weeks; seven had APC and six had metastatic CRC. Disease stabilization occurred in one patient at dose level 1, two patients at dose level 2, six patients at dose level 3, and four patients at dose level 4. The group of patients who achieved SD had a median of one prior therapy (range 1-5). Additionally, four patients with SD had a greater than 50% decrease in tumor markers (CA19-9/CEA) during the treatment period.

Nineteen patients were assessable for clinical benefit response using the Edmonton symptom assessment system. Overall, five

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016

Table 7
Non-hematologic toxicities.

Capecitabine (mg/m2)	1000	1250	1500	1750	Total no. of patients (Cycles) All grades n = 24 (116)
Toxicity	No. of patients (cycles) n = 6 (30)	No. of patients (cycles) n= 3 (15)	No. of patients (cycles) n= 7 (35)	No. of patients (cycles) n = 8 (36)	**Grade 3 and higher (%)**
Anorexia					
Grade 1-2	2 (2)	1 (2)	6 (6)	7 (7)	16 (17)
Grade 3-4	0				**0%**
Constipation					
Grade 1-2	5 (7)	2 (2)	8 (8)	3 (3)	15 (17)
Grade 3-4					**0%**
Fatigue					
Grade 1-2	1 (1)	2 (4)	4 (4)	5 (5)	13 (15)
Grade 3-4	1 (1)				**4%**
Diarrhea					
Grade 1-2	4 (5)	4 (8)	2 (7)	2 (6)	14 (28)
Grade 3-4				2 (2)	**8%**
Hand-Foot Syndrome					
Grade 1	1 (1)	1 (1)	3 (4)	4 (5)	12 (20)
Grade 2	1 (1)		3 (4)	2 (2)	**Grade 2 or higher–38%**
Grade 3			1 (1)	2 (2)	
Nausea					
Grade 1-2	3 (4)	1 (1)	2 (4)	3 (6)	12 (17)
Grade 3-4			1 (1)	1 (1)	**8%**
Vomiting					
Grade 1-2	3 (5)	1 (1)	2 (3)	4 (4)	10 (14)
Grade 3-4			1 (1)		**4%**
Heartburn					
Grade 1-2	3 (4)	2 (3)	2 (2)	3 (3)	10 (12)
Grade 3-4					**0%**
Dry Mouth					
Grade 1-2	3 (3)	1 (1)	3 (3)	2 (2)	9 (10)
Grade 3-4					**0%**
Hyperglycemia					
Grade 1-2			1 (1)	5 (7)	6 (12)
Grade 3-4			1 (1)	1 (3)	**8%**
Mucositis					
Grade 1-2	1 (1)		2 (2)	2 (2)	5 (6)
Grade 3-4				1 (1)	**4%**
Infection					
Grade 1-2	2 (2)			1 (2)	5 (6)
Grade 3-4				1 (1)	**4%**
AST/ALT					
Grade 1-2		1 (1)	1 (1)	1 (1)	4 (4)
Grade 3-4	1 (1)				**4%**

patients experienced a $\geq$10-point change towards symptom improvement, ten patients reported stable symptoms, and four patients reported $\geq$10-point change towards clinical worsening during the treatment period.

Twenty-two patients were evaluable for response. With a median follow-up of four months, the median progression-free, and overall survival times were 11.1 weeks, and 16.3 weeks, respectively.

Discussion

This is the first clinical study to evaluate the feasibility of the botanical formulation, PHY906, and capecitabine in patients with APC and other GI malignancies (Saif 2008). The combination of PHY906 and capecitabine investigated in this population seems to be well-tolerated and the capecitabine dose reached 1750 mg/m^2 BID without any DLT. The MTD was determined to be 1500 mg/m^2 BID for 7 days in combination with PHY906 800 mg BID on days 1-4 of a 14-day cycle. The main objective of this trial was to define a palliative chemotherapy regimen with an acceptable toxicity profile that could potentially improve the tolerability and efficacy of capecitabine.

Cartwright et al. demonstrated that capecitabine 1250 mg/m^2 BID administered in a 14/7 schedule had clinically significant beneficial effects in chemotherapy-naïve APC patients, and was relatively well-tolerated (Cartwright et al., 2002). Boeck et al. also showed capecitabine 1250 mg/m^2 BID in a 14/7 schedule to be effective in controlling disease in gemcitabine-pretreated patients. In our study we chose to use a 7/7 intermittent dosing schedule based on multiple studies. Scheithauer et al. found that a

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016

7/7 intermittent dosing (1750 mg/m^2 BID = total daily dose of 3500 mg/m^2) was just as active as a 14/7 dosing when used in combination with oxaliplatin in CRC patients (Scheithauer et al., 2003).

Recent data using mathematical modeling suggests that drug delivery beyond seven days contributes to toxicity with diminishing anticancer effects (Traina et al., 2008). Traina and colleagues achieved an MTD of capecitabine of 2000 mg BID fixed-dosing for seven consecutive days followed by a seven-day rest period in breast cancer patients (Traina et al., 2008). In this study, we used BSA-based dosing of capecitabine and have shown the MTD to be 1500 mg/m^2 BID in a 7/7 dosing schedule. Compared to the fixed-dose 7/7 used by Traina et al., the average BSA in our patients was 1.88 mg/m^2 yielding an median starting dose of 2725 mg at the MTD and for the whole study, significantly more than the 2000 mg BID fixed-dosing. Compared to conventional 14/7 capecitabine dosing of 1250 mg/m^2, our 7/7 schedule of 1500 mg/m^2 achieves a 90% relative dose intensity (1000 mg/m^2 less per week), and may be a more tolerable schedule, especially when used in conjunction with PHY906.

In traditional eastern herbal medicine, multiple herbs are used together either to induce a synergistic effect, enhance the efficacy, or reduce the side effects of the formulation. As standard practice in China, for example, combinations of multiple herbs are widely administered alone or in combination with Western pharmaceuticals as intravenous and oral preparations. PHY906 is an oral botanical formulation that has been found to enhance the therapeutic index of commonly used chemotherapeutic agents such as capecitabine and irinotecan in various tumor models (Liu et al., 2001). In a previous clinical study of PHY906 with irinotecan and 5-FU/LV in colorectal cancer, the frequency of diarrhea and vomiting was significantly lower with PHY906 than with placebo treatment (Farrell and Kummar, 2003). In a Phase I/II clinical study using PHY906 and capecitabine in a 14/7 schedule for patients with advanced hepatocellular carcinoma (HCC), there was no Grade 3 or 4 diarrhea (Yen et al., 2008), although the MTD of capecitabine achieved in that study was only 750 mg/m^2 BID. Although inter-study comparisons can be difficult to draw, in our study which achieved an MTD of 1500 mg/m^2 in a 7/7 schedule, the incidence of Grade 3-4 diarrhea was 8%, compared to 17% in the capecitabine 1250 mg/m^2 BID 14/7 schedule used by Cartwright et al., a 50% reduction. The incidence of Grade 2 or higher diarrhea in our study was 13%, identical to that in the study by Boeck (Boeck et al., 2007).

Therefore, PHY906 may be contributing to a cytoprotective antidiarrheal effect, making treatment with capecitabine more tolerable. In our study, mild constipation was observed in 15 patients, three of which were Grade 2. Nine of the 15 patients who reported constipation did not experience diarrhea during their treatment course. Three patients with baseline diarrhea did not report any further episodes during the study period (Hoimes et al., 2009). Indeed, PHY906 has been used historically to treat short-term diarrhea without deleterious effects, other than reversible constipation. Preclinical studies also reveal that co-administration of PHY906 and capecitabine in animal models does not alter the pharmacokinetic profile of capecitabine (Yen et al., 2008). Interestingly, one patient who experienced diarrhea from prior treatment with capecitabine before entry into the study, and was treated at the 1500 mg/m^2 cohort with PHY906, did not experience any diarrhea during the study. The patient discontinued the study due to non-adherence after three cycles, was treated with capecitabine alone at the discretion of the investigator, and experienced a recurrence of diarrhea.

As mentioned earlier, a similar Phase I/II study of PHY906 in combination with capecitabine in advanced HCC has been reported (Yen et al., 2008). This study used a range of PHY906 dosing from 600-1000 mg on days 1-4 and 8-11 of a 21-day cycle, and achieved an MTD of 750 mg/m^2 of capecitabine administered in a 14/7 schedule with PHY906 800 mg BID. Although the dose of capecitabine was considerably lower in this patient population, there was no Grade 3-4 diarrhea reported (Yen et al., 2008).

Beyond the cytoprotective benefit of PHY906, PHY906 also potentiates the effect of chemotherapy in preclinical models. In a preclinical tumor-bearing mouse model using PANC-1 tumors, PHY906 alone has little, if any, cytotoxic anti-tumor activity, but it potentiates the action of capecitabine when given in combination (Saif et al., 2007). Biochemical studies reveal that the PHY906 formulation possesses a wide range of pharmacological activities. We hypothesize that the potential mechanism(s) of action of PHY906 may include (1) enhancement of cellular uptake of chemotherapeutic agents via inhibition of multi-drug resistance (MDR) mechanisms; (2) modulation of NF-kB activity; (3) inhibition of matrix metalloproteinase (MMP) activity; and (4) inhibition of angiogenesis (Liu et al., 2003; Liu et al., 2004). Correlative studies including measurement of chemokine (IL-2, IL-4, IL-5, IL-10, TNF-a, IFN-g) levels, as surrogates for NF-kB expression, to further elucidate the effects of PHY906 will be performed in an ongoing Phase II trial.

In conclusion, the combination of PHY906 and capecitabine in patients with APC and other GI malignancies was well tolerated. Our results suggest that PHY906 can increase the therapeutic index of capecitabine in patients by reducing side effects such as diarrhea. The National Comprehensive Cancer Network guidelines for APC recommends capecitabine as second-line treatment and the prospective Phase II study by Boeck et al. demonstrates that capecitabine has a modest disease control rate (39%) (Boeck et al., 2007). Although the number of patients in this study is limited, PHY906 with capecitabine resulted in a disease control rate of 58% with one PR and thirteen SD. An ongoing Phase II study of PHY906 800 mg BID days 1-4 and capecitabine 1500 mg/m^2 BID days 1-7 of a 14-day cycle is underway to assess the clinical activity and tolerability of the combination in patients with gemcitabine-refractory pancreatic cancer.

Funding

This study was approved and funded by the National Comprehensive Cancer Network (NCCN) from general research support provided by Roche Laboratories.

References

Boeck, S., Wilkowski, R., Bruns, C.J., et al., 2007. Oral capecitabine in gemcitabine-pretreated patients with advanced pancreatic cancer. Oncology 73 (3–4), 221–227.

Burris 3rd, H.A., Moore, M.J., Andersen, J., et al., 1997. Improvements in survival and clinical benefit with gemcitabine as first-line therapy for patients with advanced pancreas cancer: a randomized trial. J. Clin. Oncol. 15 (6), 2403–2413.

Cartwright, T.H., Cohn, A., Varkey, J.A., et al., 2002. Phase II study of oral capecitabine in patients with advanced or metastatic pancreatic cancer. J. Clin. Oncol. 20 (1), 160–164.

Davison, S.N., Jhangri, G.S., Johnson, J.A., 2006. Cross-sectional validity of a modified Edmonton symptom assessment system in dialysis patients: a simple assessment of symptom burden. Kidney Int. 69 (9), 1621–1625.

Farrell, M.P., Kummar, S., 2003. Phase I/IIA randomized study of PHY906, a novel herbal agent, as a modulator of chemotherapy in patients with advanced colorectal cancer. Clin. Colorectal Cancer 2 (4), 253–256.

Hoimes C., Podoltsev N., Lamb L., Ruta S., Mezes M., Elligers K., Grant N., Liu S., Cheng Y., Saif M.W. Incidence of diarrhea in patients with GI malignancies treated with PHY906 and capecitabine (CAP) 7/7. 2009 Gastrointestinal Cancers Symposium; Abstract # 172.

Jemal, A., Siegel, R., Ward, E., Murray, T., Xu, J., Thun, M.J., 2007. Cancer statistics, 2007. CA Cancer J. Clin. 57 (1), 43–66.

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016

Liu, S.-H., Jiang, Z., Cheng, Y.-C.A., 2001. Chinese medicine formulation, PHY-906, can enhance the therapeutic Index of CPT-11 and other anticancer drugs against cancer in mice. Proc. Am. Assoc. Cancer Res. 42 (85), 458.

Liu S.-H., Jiang Z., Gao W., et al., PHY906, a Chinese herbal formulation enhances the therapeutic effect of cancer chemotherapy in human colorectal and liver cancer. In: Proc. Am. Soc. Clin. Oncol. 2003;Abstr #864.

Liu, S.-H., Jiang, Z., Leung, D., Lee, Y., Cheng, Y., 2004. PHY906: Enhancement of Cancer Chemotherapeutic Agents and Insights into Mechanism of Action. Proc. "Med. 21st Century Tri-Conf. Bio-Forum, 77.

Milella, M., Gelibter, A., Di Cosimo, S., et al., 2004. Pilot study of celecoxib and infusional 5-fluorouracil as second-line treatment for advanced pancreatic carcinoma. Cancer 101 (1), 133–138.

Mitry, E., Ducreux, M., Ould-Kaci, M., et al., 2006. Oxaliplatin combined with 5-FU in second line treatment of advanced pancreatic adenocarcinoma. Results of a Phase II trial. Gastroenterol. Clin. Biol. 30 (3), 357–363.

Moro, C., Brunelli, C., Miccinesi, G., et al., 2006. Edmonton symptom assessment scale: Italian validation in two palliative care settings. Support Care Cancer 14 (1), 30–37.

National Cancer Institute U.S. National Institute of Health. Common Terminology Criteria for Adverse Events v3.0 (CTCAE). (Publish date August 9, 2006).

Neuhaus, P., Riess, H., Post, S., et al., 2008. CONKO-1: Final results of the randomized, prospective, multicenter Phase III trial of adjuvant chemotherapy with gemcitabine versus observation with resected pancreatic cancer. JCO 26 (May 20 suppl, LBA4504).

Saif, M.W., Liu, S., Elfiky, A., Jiang, Z., Cheng, Y., 2007. Synergistic activity of PHY906 with capecitabine in pancreatic carcinoma. J. Clin. Oncol. 25 (18S), 15116.

Saif, M.W., 2008. Is there a role for herbal medicine in the treatment of pancreatic cancer? Highlights from the "44th ASCO Annual Meeting". Chicago, IL, USA. May 30–June 3, 2008. JOP 9 (4), 403–407.

Scheithauer, W., Kornek, G.V., Raderer, M., et al., 2003. Randomized multicenter Phase II trial of two different schedules of capecitabine plus oxaliplatin as first-line treatment in advanced colorectal cancer. J. Clin. Oncol. 21 (7), 1307–1312.

Therasse, P., Arbuck, S.G., Eisenhauer, E.A., et al., 2000. New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. J. Natl. Cancer Inst. 92 (3), 205–216.

Traina, T.A., Theodoulou, M., Feigin, K., et al., 2008. Phase I study of a novel capecitabine schedule based on the Norton-Simon mathematical model in patients with metastatic breast cancer. J. Clin. Oncol. 26 (11), 1797–1802.

Tsavaris, N., Kosmas, C., Skopelitis, H., et al., 2005. Second-line treatment with oxaliplatin, leucovorin and 5-fluorouracil in gemcitabine-pretreated advanced pancreatic cancer: a Phase II study. Invest. New Drugs 23 (4), 369–375.

Vignaroli, E., Pace, E.A., Willey, J., Palmer, J.L., Zhang, T., Bruera, E., 2006. The Edmonton Symptom Assessment System as a screening tool for depression and anxiety. J. Palliat. Med. 9 (2), 296–303.

Yen, Y., So, S., Rose, M., et al., 2008. Phase I/II study of capecitabine and PHY906 in hepatocellular carcinoma. J. Clin. Oncol. 26 (May 20 suppl; abstr 4610).

Please cite this article as: Saif, M.W., et al., Phase I study of the botanical formulation PHY906 with capecitabine in advanced pancreatic and other gastrointestinal malignancies. Phytomedicine (2010), doi:10.1016/j.phymed.2009.12.016