Protocol Body

1.0 Objectives

1.1 The primary objective of this study is to evaluate the efficacy of a combination of capecitabine and radiation therapy with or without curcumin in locally advanced rectal cancer as assessed by pathological complete response rate.

1.2 Secondary objectives include:
- 1.2.1 To determine downstaging, local control, disease-free survival and overall survival rates.
- 1.2.2 To determine serum and rectal tumor tissue pharmacology of curcumin and its metabolites in the above patients and its correlation with clinical response.
- 1.2.3 To identify surrogate molecular markers for curcumin effects.
- 1.2.4 To correlate serum cytokine levels with quality of life in patients receiving this therapy.

2.0 Background

Rectal cancer: Rectal cancer occurs in about 35,000 patients in the United States per year, accounting for about 30% of the cancers in the large bowel (1). Surgery is the most important component of treatment for rectal cancer and generally entails removal of the involved rectum with margins and with its vascular supply and lymphatic drainage. Standard treatment for early stage cancers is surgical resection alone. However, most rectal cancers seen in the clinic are locally advanced and some are metastatic. The German rectal cancer trial, along with multiple Phase II clinical trials, has established pre-operative chemoradiation therapy (CRT) as the standard of care for locally advanced rectal cancer patients (2-4). Compared to post-operative chemoradiation, pre-operative chemoradiation increases local control and sphincter preservation rates, and is associated with lower rates of acute and late toxicity. In the German rectal cancer trial, 8% of patients treated with pre-operative chemoradiation had a pathological complete response (pCR), and the 5-year local recurrence rate in patients treated with preoperative chemoradiation was 6%. The 5-year disease-free survival (DFS) and overall survival (OS) rates were 68% and 76%. By comparison, over 500 rectal cancer patients have been treated at M.D. Anderson Cancer Center with preoperative chemoradiation in the past decade. In this series, the pathologic complete response rate was 19% with downstaging rate of 57%, and 5-year local recurrence rates of 10%; the corresponding 5-year DFS and OS survival rates were 79% and 80%, respectively (3, 5). We and other groups have recognized that response to pre-operative CRT is an excellent predictor of better DFS and OS rates (6-10). The median DFS and overall survival rates for responders were 46 months and 47 months, respectively; for non-responders these outcome measures were 38 months and 41 months, respectively (6). In addition, response to pre-operative CRT may improve the chances of sphincter-preserving surgery (11, 12). Lastly, a complete pathological response to CRT may permit selected patients to undergo less extensive resections, further minimizing toxicity (13).

Our goal is to further increase the pCR rate and downstaging rate, which we will use as surrogates for overall clinical outcomes. Strategies include increasing the dose of radiation without increasing surgical morbidity, and optimizing systemic therapy during radiation. Building on the radiobiological advantage documented in head and neck cancer, we have increased the intensity of radiation therapy by delivering two doses of radiation therapy (concomitant boost) during the last week of the pre-operative regimen (4, 14, 15). This resulted

in a pCR rate of 18%, no different from that of prior experiences. We are presently investigating the combination of standard fluoropyrimidine-based chemotherapy with anti-angiogenic therapy using Bevacizumab. Other groups have combined newer targeted agents to the cytotoxic backbone of radiation therapy and fluoropyrimidine-based chemotherapy (16-18). No regimen has successfully improved overall outcomes and many have increased toxicity. Most strategies that target radioresistance of tumors have focused on the inherently or constitutively overexpressed pro-survival pathways within tumor cells (19, 20). Multiple signaling pathways and redundancies, however, make it difficult to overcome the blockade of a specific pathway (21, 22). An alternative approach that has interested us is broad-spectrum blockade of inducible pathways that transiently drive pro-survival and anti-apoptotic signals to overcome the cytotoxic signal triggered by treatment. An agent of particular interest in this regard is curcumin.

Curcumin: Curcumin (diferuloylmethane) is a polyphenol, and is the major constituent of the yellow spice turmeric, derived from the rhizomes of Curcuma longa. The medicinal use of this plant has been documented in Ayurveda, the Indian system of medicine, for centuries (23, 24). It is commonly used as a spice or flavoring agent, and has been widely utilized in India and southeast Asia mainly for its anti-inflammatory properties. While the anti-inflammatory properties of curcumin have been demonstrated in the scientific literature decades ago, more recently curcumin has shown potential anti-neoplastic and chemopreventive promise in a number of in vitro, in vivo and clinical studies (25, 26). Curcumin has been shown to inhibit the proliferation of a wide variety of tumor cells (27-34). It inhibits angiogenesis, induces apoptosis or cell cycle arrest, and causes regression of tumors in preclinical models (28-32, 35-37).

Mechanism of anti-tumor activity of curcumin: Although the exact mechanisms for its anti-tumor properties are not fully understood, extensive research indicates that curcumin inhibits a number of molecular pathways that contribute to cell survival, proliferation, invasion, and metastasis of cancer cells. This includes inhibition of signal transduction pathways and tumor angiogenesis and induction of apoptosis (27-34). Studies from our group and others have shown that activation of nuclear factor kappa B (NF-kB) promotes cell survival and cell proliferation, whereas downregulation of NF-kB sensitizes cells to apoptosis (38-44). Thus, suppression of NF-kB in cancer cells may provide a molecular target for prevention and treatment of cancer (40, 45-49). Our group and others have shown that curcumin is a potent down regulator of NF-kB activation, most likely through suppression of inhibitor of NF-kB (IkB) kinase activity, and inhibition of NF-kB mediated gene expression (25, 27-30, 32, 36, 50-55). In keeping with these proliferative signals induced by NF-kB, constitutive activation of NF-B is found in many malignancies including colorectal cancer (56, 57). NF-kB regulated genes include bcl-2, cyclo-oxygenase-2 (COX2), matrix-metalloproteinase-9 (MMP-9), and adhesion molecules such as ICAM-1 and VCAM-1 (58). Since NF-kB regulates several genes that are involved in cellular transformation, proliferation, invasion, and angiogenesis, suppression of NF-kB activation may be one of the mechanisms of actions by which curcumin mediates its anti-tumor effects. Additionally, it inhibits induction of COX2, an enzyme implicated in colorectal carcinogenesis (59-61). This is best explained by inhibition of IkB kinase (IKK) activity which is responsible for IkB phosphorylation (54).
In addition, curcumin affects other signaling mechanisms that are critical for tumor growth including inhibition of epidermal growth factor receptor (EGFR) signaling and tyrosine kinase activity and downregulation of the transcription factor c-Jun/AP-1 and STAT3, which may account for the inhibitory effects of curcumin on proliferation of various tumor cell lines (62-67). Specifically in colorectal cancer, curcumin stimulates the transactivating activity of peroxisome proliferator-activated receptor which appears to mediate suppression of EGFR and cyclin D1 genes (68). Curcumin also has immunomodulatory effects mediated via activation of host

macrophages and natural killer cells and modulation of lymphocyte-mediated function (69, 70). Therefore, the exact mechanism of action of curcumin remains inadequately explained. However, the most consistent finding is that curcumin induces apoptosis. This may be mediated by impairment of the ubiquitin proteasome pathway (71), upregulation of proto-oncoprotein Bax (72, 73), activation of caspases and induction of Fas receptor aggregation in a FasL dependent manner (73), and the generation of free radicals (74).

The NF-kB pathway:
NF-kB resides in the cytoplasm of cells in the inactive state in a complex with inhibitors of kB (IkBs). NF-kB is activated by phosphorylation of IkB-a at two serine residues (Ser-32 and –36) or, less frequently, at a tyrosine residue (Tyr-42) by IkB kinases leading to its ubiquitination and degradation by the proteosome complex. This leads to release of the active NF-kB, which translocates to the nucleus and binds to its consensus sequence in the promoter or enhancer region. This further leads to transcription of a wide variety of anti-apoptotic genes and other pleiotropic effects. The usual triggers for NF-kB activation are cytokines such as IL-1 and TNF-a, bacterial or viral infections, growth factors acting via the atypical kinase (Akt) pathway, and stress (including reactive oxygen intermediates and ionizing radiation). The effector proteins include acute phase reaction proteins, cytokines, adhesion molecules and feedback regulatory proteins that moderate the NF-kB response. In colorectal cancer in particular, cyclo-oxygenase-2 (cox-2) is a key mediator of tumor progression whose expression is also mediated by NF-kB.

Targeting *inducible*- rather than *constitutive*- pro-survival pathways as a means to enhance radioresponse: Tumors that constitutively overexpress pro-survival signaling molecules are well characterized as more resistant to radiation therapy than those with normal or low expression levels. This is a classical target for radiosensitization strategies including newer therapies that target epidermal growth factor receptor, restore p53, block bcl-2, and target Ras. The efficacy of highly targeted therapy may be diminished by up-regulation of redundant signaling pathways that signal downstream of the initial targeting and overcome such blockade. For instance, in a study of gliomas cells using a targeted inhibitor of the EGFR pathway, cells upregulated insulin-like growth factor receptor-1, which then triggered signaling downstream (PI3kinase/Akt and Ras/MAPK pathways) from the initial EGFR inhibition to overcome this blockade (21). An alternative strategy to enhance radioresponse is to target inducible radioresistance pathways. These are often transiently up-regulated in response to radiation therapy and can provide the necessary stimuli to increase proliferation and inhibit apoptosis. This response is very similar to classical radiobiology principles of radioresistance where repopulation (proliferation of tumor cell clonogens between two fractions of sublethal radiation therapy) leads to decrease in effectiveness of a course of radiation therapy. This is illustrated by the cartoon in Figure 1 showing the theoretical effect of tumor repopulation on tumor control probability after a 30-day course of fractionated radiation therapy. In each curve, 50% cell killing was assumed for each dose of radiation. Continued cell proliferation (50% re-growth was assumed) results in partial tumor repopulation between fractions of radiation (light line). Pharmacologic inhibition of tumor repopulation (dark line) could significantly improve the efficacy of therapy.
We have demonstrated in preclinical studies that NF-kB provides one such pro-survival, anti-apoptotic signal that is induced transiently by radiation therapy and protects cells from radiation-induced apoptosis (see preliminary results) (75). Colorectal cancers are known to have low baseline constitutive **NF-kB activity** and our preclinical studies have shown that this **is further activated by radiation therapy**. Targeting this radiation-induced pro-survival signaling pathway is a novel method to potentiate radioresponse. We have demonstrated that **curcumin inhibits this radiation-induced *NF-kB activation*** and increases radioresponse of colorectal

cancer cells. ***Consequently, our proposal to use a non-toxic broad-spectrum agent, curcumin, to target an inducible radioresistance pathway, the NF-kB pathway, is both novel and logical.***

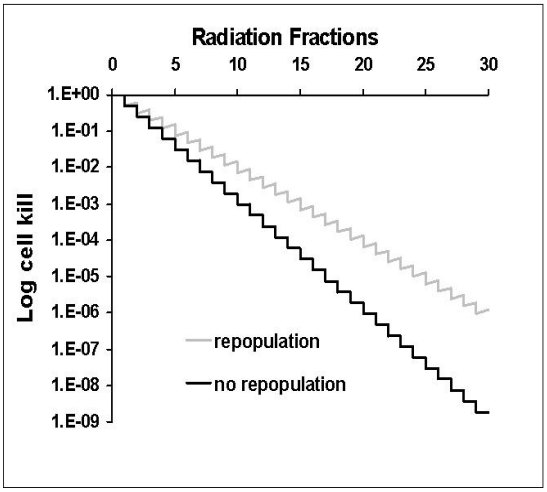


Fig. 1: Theoretical effect of tumor re-population on tumor control probability.

Epidemiological significance: The higher prevalence of colorectal cancer in the Western world is attributed to dietary factors including higher intake of animal fat and lower intake of fiber. This is supported by the finding that Japanese immigrants to the United States have a greater than 2-fold increase in the development of colorectal cancer than native Japanese. Decreased prevalence in Asian and African countries where fiber and cellulose are consumed in greater quantities also supports the role of dietary factors in colorectal carcinogenesis. Within India, the incidence of colorectal cancer is higher in Northern India than Southern India. One possible explanation is the increased consumption of curry-containing foods in South India, where the coloring agent turmeric is used.

Capecitabine: A randomized trial by the Gastrointestinal Intergroup established protracted venous infusional 5-FU as the standard concurrent chemotherapy with radiation (76). Oral capecitabine pharmacologically mimics protracted venous infusional 5-FU. A recent phase III trial on adjuvant therapy for stage III colon cancer showed that capecitabine gave equivalent disease-free survival as bolus 5-FU/leukovorin, with a trend towards improved disease-free survival and overall survival, as well as decreased neutropenia and stomatitis (77). Capecitabine is well tolerated with radiotherapy based on data from a phase I dose escalation trial in patients with rectal cancer (78). Capecitabine was administered continuously (7 days per week) throughout a 38-day course of radiotherapy (50.4 Gy in 28 fractions). The dose limiting toxicity was hand foot syndrome, and there was significantly less gastrointestinal and hematologic toxicity than expected from infusional 5-FU. At M.D. Anderson, capecitabine has been studied in a phase II trial in patients with rectal cancer treated with neoadjuvant therapy. Our results indicate that therapy is very well tolerated and the treatment effect is comparable to 5-FU (15).

MD Anderson Symptom Inventory (MDASI) and Brief Fatigue Inventory (BFI)
The MDASI is a simple, reliable, and patient self-reported, clinically validated tool for the assessment of symptom burden and symptom clusters in patients undergoing treatment for cancer. Using serial administration of the MDASI during a course of RT for rectal cancer, we are able to

generate plots that represent the severity and temporal profile of individual components of a symptom cluster. This has been done on prior novel biological agent and/or radiation fractionation protocols of chemoradiation for rectal cancer and is also being performed on a separate quality of life protocol on all patients treated with chemoradiation for rectal cancer. The MDASI - gastrointestinal module (MDASI-GI) is an extension of the MDASI with questions specifically addressing GI symptoms experienced by cancer patients during chemotherapy and/or radiaiton therapy. The additional questions are being validated in an on-going collection of questionnaires during chemoradiation for rectal cancer. Similarly, the BFI was developed for the rapid assessment of fatigue severity for use in both clinical screening and clinical trials. The BFI has been shown to be simple, completable by most patients, and an internally stable (reliable) measure of the severity of fatigue that correlates highly with similar fatigue measures. Both the MDASI and BFI questionnaires will be given to the patients by the research nurse and will be filled out by the patient and/or their caregiver. These questionnaires are filled weekly during radiation treatment and the completed questionnaires are handed to the research nurse during weekly treatment visits with the treating radiation oncologist.

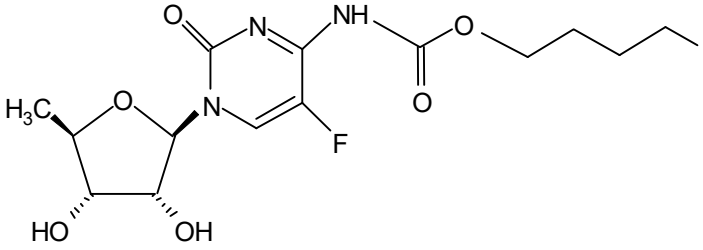
3.0 Drug Background

3.1 Capecitabine

3.1.1 Description

Capecitabine is a fluoropyrimidine carbamate with antineoplastic activity. It is an orally administered systemic prodrug of 5'-deoxy-5-fluorouridine (5'-DFUR) which is converted to 5-fluorouracil.

The chemical name for capecitabine is 5'-deoxy-5-fluoro-N-[(pentyloxy) carbonyl]-cytidine and has a molecular weight of 359.35. Capecitabine has the following structural formula:



Capecitabine is a white to off-white crystalline powder with an aqueous solubility of 26 mg/mL at 20ºC.

Capecitabine is supplied as biconvex, oblong film-coated tablets for oral administration. Each light peach-colored tablet contains 150 mg capecitabine and each peach-colored tablet contains 500 mg capecitabine. The inactive ingredients in capecitabine include: anhydrous lactose, croscarmellose sodium, hydroxypropyl methylcellulose, microcrystalline cellulose, magnesium stearate and purified water. The peach or light peach film coating contains hydroxypropyl methylcellulose, talc, titanium dioxide, and synthetic yellow and red iron oxides.

Drug Formulation, Packaging, and Storage

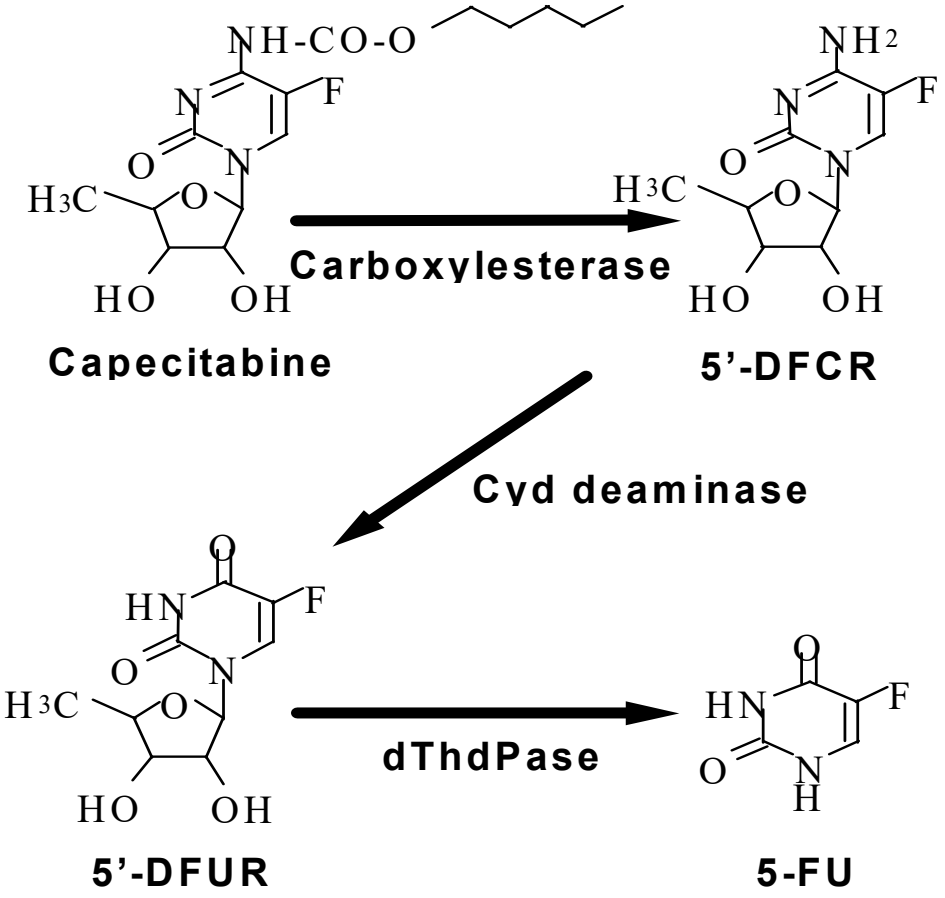
Capecitabine is commercially available as 150 mg or 500 mg tablets. Capecitabine tablets are packed in polyethylene bottles, containing either 120 x 150 mg tablets or 240 x 500 mg tablets. Capecitabine tablets should be stored at room temperature (15 to 30C) in the container in which they are provided.

3.1.2 Clinical Pharmacology

Capecitabine is relatively non-cytotoxic in vitro. This drug is enzymatically converted to 5-fluorouracil (5-FU) in vivo.

Bioactivation: Capecitabine is readily absorbed from the gastrointestinal tract. In the liver, a 60 kDa carboxylesterase hydrolyzes much of the compound to 5'-deoxy-5-fluorocytidine (5'-DFCR). Cytidine deaminase, an enzyme found in most tissues, including tumors, subsequently converts 5'-DFCR to 5'-deoxy-5-fluorouridine (5'-DFUR). The enzyme, thymidine phosphorylase (dThdPase), then hydrolyzes 5'-DFUR to the active drug 5-FU. Many tissues throughout the body express thymidine phosphorylase. Some human carcinomas express this enzyme in higher concentrations than surrounding normal tissues.

Metabolic Pathway of capecitabine to 5-FU



3.1.3 Mechanism of Action:

Both normal and tumor cells metabolize 5-FU to 5-fluoro-2'-deoxyuridine monophosphate (FdUMP) and 5-fluorouridine triphosphate (FUTP). These metabolites cause cell injury by two different mechanisms. First, FdUMP and the folate cofactor, N5-10-methylenetetrahydrofolate, bind to thymidylate synthase (TS) to form a covalently bound ternary complex. This binding inhibits the formation of thymidylate from 2'-deoxyuridylate. Thymidylate is the necessary precursor of thymidine triphosphate, which is essential for the synthesis of DNA, so that a deficiency of this compound can inhibit cell division. Second, nuclear transcriptional enzymes can mistakenly incorporate FUTP in place of uridine triphosphate (UTP) during the synthesis of RNA. This metabolic error can interfere with RNA processing and protein synthesis.

3.1.4 Pharmacokinetics in Colorectal Tumors and Adjacent Healthy Tissue:

Following oral administration of capecitabine 7 days before surgery in patients with colorectal cancer, the median ratio of 5-FU concentration in colorectal tumors to adjacent tissues was 2.9 (range from 0.9 to 8.0). These ratios have not been compared to 5-FU infusion.

3.1.5 Human Pharmacokinetics:

The pharmacokinetics of capecitabine and its metabolites have been evaluated in about 200 cancer patients over a dosage range of 500 to 3500 mg/m2/day. Over this range, the pharmacokinetics of capecitabine and its metabolite, 5'-DFCR were dose proportional and did not change over time. The increases in the AUCs of 5'-DFUR and 5-FU, however, were greater than proportional to the increase in dose and the AUC of 5-FU was 34% higher on day 14 than on day 1. The elimination half-life of both parent capecitabine and 5-FU was about ¾ of an hour. The inter-patient variability in the Cmax and AUC of 5-FU was greater than 85%.

3.1.6 Absorption, Distribution, Metabolism and Excretion:

Capecitabine reached peak blood levels in about 1.5 hours (Tmax) with peak 5-FU levels occurring slightly later, at 2 hours. Food reduced both the rate and extent of absorption of capecitabine with mean Cmax and AUC decreased by 60% and 35%, respectively. The Cmax and AUC(0-infinity) of 5-FU were also reduced by food by 43% and 21%, respectively. Food delayed Tmax of both parent and 5-FU by 1.5 hours. Plasma protein binding of capecitabine and its metabolites is less than 60% and is not concentration-dependent. Capecitabine was primarily bound to human albumin (approximately 35%). Capecitabine is extensively metabolized enzymatically to 5-FU. The enzyme dihydropyrimidine dehydrogenase hydrogenates 5-FU, the product of capecitabine metabolism, to the much less toxic 5-fluoro-5, 6-dihydro-fluorouracil (FUH2). Dihydropyrimidinase cleaves the pyrimidine ring to yield 5-fluoro-beta-ureido-propionic acid (FUPA).

Finally, beta-ureido-propionase cleaves FUPA to alpha-fluoro-beta-alanine (FBAL) which is cleared in the urine. Capecitabine and its metabolites are predominantly excreted in urine; 95.5% of administered capecitabine dose is recovered in urine. Fecal excretion is minimal (2.6%). The major metabolite excreted in urine is FBAL which represents 57% of the administered dose. About 3% of the administered dose is excreted in urine as unchanged drug.

3.1.7 Capecitabine Drug-Drug Interactions:WARNING

3.1.7.1 Coumarin (Coumadin):

Concurrent use of coumadin other than low dose (1 mg) coumadin used for line patency is contraindicated. Patients on coumadin must be changed to Lovenox at least 1 week prior to starting capecitabine.

Altered coagulation parameters and/or bleeding, including death, have been reported in patients taking capecitabine concomitantly with coumarin-derivative anticoagulants such as warfarin and phenprocoumon. These events occurred within several days and up to several months after initiating capecitabine therapy and, in a few cases, within one month after stopping capecitabine and occurred in patients with and without liver metastases. In a drug interaction study with single dose warfarin administration, there was a significant increase in the mean AUC of S-warfarin. The maximum observed INR value increased by 91%. This interaction is probably due to an inhibition of cytochrome P450 2C9 by capecitabine and/or its metabolites.

3.1.7.2 Phenytoin

Increased phenytoin plasma concentrations have been reported during concomitant use of capecitabine with phenytoin, suggesting a potential interaction. Patients taking phenytoin concomitantly with capecitabine should be regularly monitored (e.g. weekly phenytoin and albumin levels) for increased phenytoin plasma concentrations and associated clinical symptoms.

3.1.8 Clinical Studies

Colorectal Cancer
The efficacy and safety of three capecitabine dosing schedules, when administered as first-line treatment, was evaluated in 109 patients with advanced colorectal cancer. The three dosing schedules were as follows: capecitabine 2510 mg/m2/day intermittent (2 weeks with treatment, 1 week without treatment), capecitabine 1657 mg/m2/day intermittent in combination with leucovorin 60 mg/day, capecitabine 1331 mg/m2/day continuous (no rest period). Overall response rates were 24%, 23% and 21% in the three groups, respectively. Thirteen percent of the patients discontinued treatment due to treatment-related adverse events (79).

Two large randomized phase III studies evaluated further the efficacy of capecitabine as first line treatment in metastatic colorectal cancer. Capecitabine was administered at a dose of 2500 mg/m2/day, and was compared with standard treatment (5-FU at a dose of 425 mg/m2 in combination with leucovorin 20 mg/m2 days 1-5 every 4 weeks). In the North American trial, 24.8% in the capecitabine group had a response, compared with 15.5% in the 5-FU/leucovorin group

(p=0.005). Related serious adverse events and the rate of hospitalizations was significantly lower (p<0.05) in the capecitabine group (80). The European study demonstrated responses in 18.9% of capecitabine patients and 15.0% of the 5-FU/leucovorin patients (16.1%). Capecitabine resulted in significantly lower incidences of stomatitis, alopecia and grade 3-4 neutropenia, but a higher incidence of hand-foot syndrome (81).

A recent phase III trial on adjuvant therapy for stage III colon cancer showed that capecitabine (1,250 mg/m^2 PO BID on days 1-14 for 21 days) gave equivalent disease-free survival as bolus 5-FU/leukovorin, with a trend towards improved disease-free survival and overall survival, as well as decreased neutropenia and stomatitis (77). Capecitabine is well tolerated with radiotherapy based on data from a phase I dose escalation trial in patients with rectal cancer [16]. Capecitabine was administered continuously (7 days per week) throughout a 38-day course of radiotherapy (50.4 Gy in 28 fractions). The dose limiting toxicity was hand foot syndrome, and there was significantly less gastrointestinal and hematologic toxicity than expected from infusional 5-FU (78). At M.D. Anderson, capecitabine has been studied in a phase II trial in patients with rectal cancer treated with neoadjuvant therapy. Our results indicate that the therapy is very well tolerated and the treatment effect is comparable to 5-FU (15). No randomized data currently exists comparing concurrent capecitabine and radiotherapy with concurrent continuous infusion 5-FU and radiotherapy for rectal cancer. However, since the X-ACT trial showed that capecitabine is at least equivalent to 5FU/ and leucovorin for the adjuvant treatment of colon cancer, concurrent capecitabine and radiotherapy has been accepted as standard of care for preoperative treatment of rectal cancer at many institutions, including M.D. Anderson.

For more than 7 years now, at M.D. Anderson, capecitabine has been typically given at a dose of 825 mg/m^2, Monday to Friday, concurrently with radiotherapy for preoperative treatment of rectal cancer.

Adverse Reactions
A total of 596 patients with metastatic colorectal cancer were treated with 1250 mg/m^2 twice a day of capecitabine administered for 2 weeks followed by a 1-week rest period, and 593 patients were administered 5-FU and leucovorin in the Mayo regimen (20 mg/m^2 leucovorin IV followed by 425 mg/m^2 IV bolus 5-FU, on days 1-5, every 28 days). In the pooled colorectal database the median duration of treatment was 139 days for capecitabine-treated patients and 140 days for 5-FU/LV treated patients. A total of 78 (13%) and 63 (11%) capecitabine and 5-FU/LV-treated patients, respectively, discontinued treatment because of adverse events/intercurrent illness. A total of 82 deaths due to all causes occurred either on study or within 28 days of receiving study drug: 50 (8.4%) patients randomized to capecitabine and 32 (5.4%) randomized to 5-FU/LV.

3.1.9 Special Populations:

A population analysis of pooled data from the two large controlled studies in patients with colorectal cancer (n=505) who were administered Capecitabine at

1250 mg/m2 twice a day indicated that gender (202 females and 303 males) and race (455 white/caucasian patients, 22 black patients, and 28 patients of other race) have no influence on the pharmacokinetics of 5'-DFUR, 5-FU and FBAL. Age has no significant influence on the pharmacokinetics of 5'-DFUR and 5-FU over the range of 27 to 86 years. A 20% increase in age results in a 15% increase in AUC of FBAL.

3.2.9.1 Hepatic Insufficiency:

Capecitabine has been evaluated in 13 patients with mild to moderate hepatic dysfunction due to liver metastases defined by a composite score including bilirubin, AST/ALT and alkaline phosphatase following a single 1255 mg/m^2 dose of capecitabine. Both AUC(0-infinity) and C_{max} of capecitabine increased by 60% in patients with hepatic dysfunction compared to patients with normal hepatic function (n=14). The AUC(0-infinity) and C_{max} of 5-FU was not affected. In patients with mild to moderate hepatic dysfunction due to liver metastases, caution should be exercised when capecitabine is administered. The effect of severe hepatic dysfunction on capecitabine is not known .

3.2.9.2 Renal Insufficiency*:*

Capecitabine is contraindicated in patients with severe renal impairment (creatinine clearance below 30 mL/min [Cockroft and Gault]). In patients with moderate renal impairment (creatinine clearance 30-50 mL/min [Cockroft and Gault]) at baseline, a dose reduction to 75% of the Capecitabine starting dose is recommended. In patients with mild renal impairment (creatinine clearance 51-80 mL/min) no adjustment in starting dose is recommended.

Cockroft and Gault Equation:

$$\textit{Creatinine clearance for males} = \frac{\textit{(140 - age [yrs]) (body wt [kg])}}{\textit{(72) (serum creatinine [mg/dL])}}$$

Creatinine clearance for females = 0.85 x male value

Following oral administration of 1250 mg/m^2 capecitabine twice a day to cancer patients with varying degrees of renal impairment, patients with moderate (creatinine clearance = 30-50 mL/min) and severe (creatinine clearance <30 mL/min) renal impairment showed 85% and 258% higher systemic exposure to FBAL on day 1 compared to normal renal function patients (creatinine clearance >80 mL/min). Systemic exposure to 5'-DFUR was 42% and 71% greater in moderately and severely renal impaired patients, respectively, than in normal patients. Systemic exposure to capecitabine was about 25% greater in both moderately and severely renal impaired patients.

3.2.9.3 ***Hand-Foot Syndrome****:* Hand-foot syndrome (palmar-plantar erythrodysesthesia or chemotherapy induced acral erythema) is a cutaneous toxicity (median time to onset of 79 days, range from 11 to 360 days) with a severity range of grades 1 to 3. Grade 1 is characterized by any of the following: numbness, dysesthesia/paresthesia, tingling,

painless swelling or erythema of the hands and/or feet and/or discomfort which does not disrupt normal activities. Grade 2 hand-and-foot syndrome is defined as painful erythema and swelling of the hands and/or feet and/or discomfort affecting the patient's activities of daily living. Grade 3 hand-and-foot syndrome is defined as moist desquamation, ulceration, blistering or severe pain of the hands and/or feet and/or severe discomfort that causes the patient to be unable to work or perform activities of daily living. If grade 2 or 3 hand-and-foot syndrome occurs, administration of capecitabine should be interrupted until the event resolves or decreases in intensity to grade 1. Following grade 3 hand-and-foot syndrome, subsequent doses of capecitabine should be decreased.

3.2.9.4 ***Cardiotoxicity:*** The cardiotoxicity observed with capecitabine includes myocardial infarction/ischemia, angina, dysrhythmias, cardiac arrest, cardiac failure, sudden death, electrocardiographic changes, and cardiomyopathy. These adverse events may be more common in patients with a prior history of coronary artery disease.

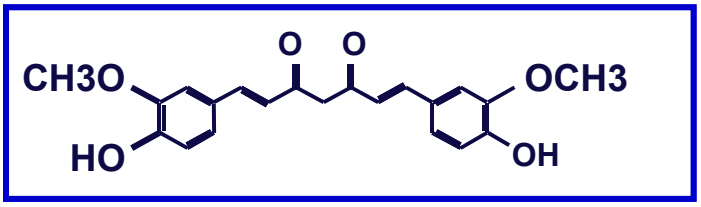
3.2.9.5 ***Hepatic Insufficiency:*** Patients with mild to moderate hepatic dysfunction due to liver metastases should be carefully monitored when capecitabine is administered. The effect of severe hepatic dysfunction on the disposition of capecitabine is not known.

3.3 Curcumin

3.3.1 Description

Curcumin (Diferuloylmethane) is a naturally occuring polyphenol, the active ingredient in the dried ground rhizome of the perennial herb *Curcuma longa*, commonly known as turmeric. Turmeric has universal appeal as a colouring agent, food preservative and flavouring agent in foods. Over the past three decades, it has been studied as an antioxidant, anti-inflammatory, cancer chemopreventive and potentially as a chemotherapeutic agent.

The chemical name for curcumin is [1,7-bis-(4-hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione]. This bis-unsaturated-diketone exists in equilibrium with its enol tautomer. Curcumin has a molecular weight of 368.37 and the following structural formula:



Curcumin is an orange-yellow crystalline powder that is insoluble in water but

soluble in acetone, alcohol and glacial acetic acid.

Drug Formulation, Packaging, and Storage

Curcumin is available as 1 gm capsules of C3Complex (Sabinsa Corporation, New Jersey). This consists of standardized extract of phenolic compounds from turmeric root (Curcuma longa fam. Zingiberaceae) containing high purity curcuminoids (minimum 95% by HPLC). Curcuminoids include 2.5-6.5% bisdemethoxy-curcumin, 15-25% demethoxy- curcumin, and 70-80% curcumin. Curcuma longa is listed in the FDA' GRAS list, a list of food additives which are "Generally Recognized As Safe" by a consensus of scientific opinion. The GRAS list appears in the Code of Federal Regulations (CFR) Title 21 Parts 172, 182, 184 and 186. C3Complex should be stored at room temperature 25º C (77º F) [excursions permitted to 15-30º C (59-86º F] in the container in which they are provided.

3.3.2 Adverse Events

Incidence rates of adverse events associated with curcumin are rare. The following events may be expected with the administration of curcumin:

3.3.3.1 Gastrointestinal: Occasional mild diarrhea has been noted in some human subjects .
3.3.3.2 Hematologic: None expected.
3.3.3.3 Hepatic: None expected
3.3.3.4 Renal: None expected
3.3.3.5 Respiratory: None expected
3.3.3.6 Anaphylactic-like reactions:. No cases of anaphylactic reactions and/or angioedema have been reported in patients receiving curcumin.
3.3.3.7 Neurological: None expected
3.3.3.8 General: None expected

3.3.3 Drug Interactions: None known.

3.3.4 Contraindications: Curcumin is contraindicated in patients with hypersensitivities to curcumin.

3.3.5 Clinical studies:

In an early report from 1987, Kuttan reported on turmeric as a topical treatment for oral cancers and leukoplakia (82). This research group reported a reduction in the size of the lesions in 10% of the 62 patients treated. The study was limited by the lack of a control group, no assessment of anti-inflammatory activity and no chemical analysis of the preparation applied. Nonetheless, the results suggest the potential for antineoplastic properties.

A phase I trial evaluated the pharmacokinetics of curcumin in Taiwanese subjects with a variety of "high risk or pre-malignant" lesions such as oral leukoplakia and cervical intraepithelial neoplasia among others (83). The starting oral dose was 500 mg a day and this was escalated to 12,000 mg. No treatment related toxicity was observed. There was a dose related increase in measured plasma levels. Of

the seven patients with leukoplakia, 2 showed histologic improvement, although progression was observed in one. In total, 7 of 25 patients enrolled showed some degree of histologic improvement.

A recent phase 1 study evaluated a curcuminoid formulation in healthy volunteers using a single dose of standardized powder extract, uniformly milled curcumin. Doses were escalated from 0.5 to 12 gm and only minimal toxicity was noted and it did not appear to dose-related. The observed adverse events included diarrhea, headache, rash, and yellow stool. Low levels of curcumin were detected in the serum in 2 subjects who received >8gm (84).

A phase I study was completed at the University of Leicester in England using 15 subjects with advanced colorectal cancers (85, 86). The subjects orally consumed curcumin doses of 450 mg up to 3.6 gm daily for up to 4 months. Two patients experienced mild diarrhea, but there was no dose limiting toxicity observed. Three indices of potential pharmacological activity of curcumin were evaluated (based on preclinical data). These were GST activity, levels of a deoxyguanosine adduct (M1G) (which is related to oxidative stress and can be altered by curcumin in the diet of animals or human subjects), and prostaglandin levels as an indicator of COX-2 activity. Curcumin consumed at 3.6 gm levels generated detectable levels of parent compound and conjugates in plasma and urine, demonstrating bioavailability outside the gastrointestinal tract. The same dose caused inhibition of PGE2 production in blood leukocytes (measured ex vivo). Therefore the authors concluded that 3.6 gm was a suitable oral dose for evaluation of phase II trials. In addition, since the measurement of compliance is important in the context of intervention trials, the consistent presence of curcumin and its conjugates in urine observed in patients consuming 3.6 gm of curcumin daily is of potential relevance to the use of curcumin as a chemopreventive agent.

In another study evaluating human pharmacokinetics, twelve patients with confirmed colorectal cancer received oral curcumin at 0.45, 1.8 or 3.6 gm per diem for 7 days prior to surgery (87). Levels of agent-derived species were determined in the peripheral circulation and in colorectal tissue obtained at the time of surgical resection. The concentrations of curcumin in normal and malignant colorectal tissue of patients consuming 3.6 g daily of curcumin were 12.7 ± 5.7 and 7.7 ± 1.8 nmol/gm tissue, respectively. Curcumin sulphate and curcumin glucuronide were identified in the intestinal tissues of these patients. Trace levels of curcumin were found in the peripheral circulation. Compatible with the preclinical data presented above, these results also suggest that a daily dose of 3.6 gm curcumin achieves relatively high levels in colorectal tissue with measurable distribution of the parent drug outside the gut.

3.3.6 Dose Modification: The NCI Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0 will be used to score drug toxicity. If attributable toxicities are encountered, the drug dosage will be modified according to the following schedule (The drug may be restarted when the toxicity subsides to grade ≤ 1, but missed doses will not be made up.)
Please note that for any event which is apparent at baseline, the dose modification will apply according to the corresponding shift in toxicity grade if the investigator feels it is appropriate (e.g. if a patient has grade 1 asthenia at baseline which increases to grade 2 during treatment, this will be considered as a shift of 1 grade

and treated as a grade 1 toxicity for dose modification purposes).

Drug dose	Toxicity grade	Drug dose modification
4 gm twice daily	0 - 1	Continue 4 gm twice daily
	2	Hold until toxicity is grade 2, then resume at 2 gm twice daily
	3 - 4	Discontinue

The drug should be discontinued if toxicity of $\leq$ Grade 1 is not reached within 4 weeks.

4.0 Patient Eligibility

4.1 Inclusion Criteria

4.1.1. All patients must have clinical stage T3,4 N0,1,2 or T2N1,2 adenocarcinoma of the rectum. Patients will be clinically staged using endorectal ultrasound, pelvic CT or MRI, and physical examination.

4.1.2 Histology must be confirmed with review by the Department of Pathology at MDACC.

4.1.3 All patients must have no distant metastatic disease in the liver, peritoneum, lungs, or paraaortic lymph nodes.

4.1.4 Patients must have a performance status (Karnofsky scale) of 70% or greater.

4.1.5 Patients must be >/= 18 years of age.

4.1.6 Patients must have ANC > 1200 cells/mm3, platelets > 100,000/mm3, total serum bilirubin < 2 mg/dl, BUN <30 mg/dl, creatinine < 1.5 mg/dl or creatinine clearance > 50cc/min (estimated as calculated with Cockcroft-Gault equation).
Creatinine Clearance (men) = {(140 - age) x lean body weight (in kg.)}/ {Plasma creatinine (mg/dl) x 72}
Creatinine Clearance (women) = 0.85 x {(140 - age) x lean body weight (in kg.)}/ {Plasma creatinine (mg/dl) x 72}

4.1.7 Patients must have signed informed consent indicating that they are aware of the investigational nature of the study, and are aware that participation is voluntary. Patients must also agree to refrain from use of additional herbal supplements during the course of the study.

4.1.8 Postmenopausal woman must have been amenorrheic for at least 12 months to be considered of non-childbearing potential. Patients will agree to continue contraception for 30 days from the date of the last study drug administration. Sexually active males must practice contraception during the study.

4.2 Exclusion Criteria

4.2.1 Prior complete course up to 5 Gy of radiotherapy to the pelvis.

4.2.2 Pregnant or lactating woman. Women of childbearing potential who have not undergone a hysterectomy with either a positive or no pregnancy test at baseline. Women / men of childbearing potential not using a reliable and appropriate contraceptive method (oral, injectable, or implantable hormonal contraceptive; tubal ligation; intra-uterine device; barrier contraceptive with spermicide; or vasectomized partner).

4.2.3 Treatment for other carcinomas within the last five years, except cured

non-melanoma skin and treated in-situ cervical cancer.

4.2.4 Patients with uncontrolled intercurrent illness including, but not limited to, ongoing or requiring IV antibiotics0, cardiac disease NYHA class III or IV, unstable angina pectoris, unstable cardiac arrythmia or tachycardia (heart rate > 100 beats/minute), or psychiatric illness/ social situations that would limit compliance with the study requirements are excluded.

4.2.5 Other serious uncontrolled medical conditions that the investigator feels might compromise study participation.

4.2.6 Major surgery within 4 weeks of the start of study treatment.

4.2.7 Prior unanticipated severe reaction to fluoropyrimidine therapy or known hypersensitivity to 5–fluorouracil or capecitabine or curcumin.

4.2.8 Concurrent use of coumadin other than low dose (1 mg) coumadin used for line patency. Patients on coumadin must be changed to Lovenox at least 1 week prior to starting capecitabine.

4.2.9 Concurrent use of cimetidine, allopurinol, or aluminium hydroxide and magnesium hydroxide-containing antacids such as Maalox.

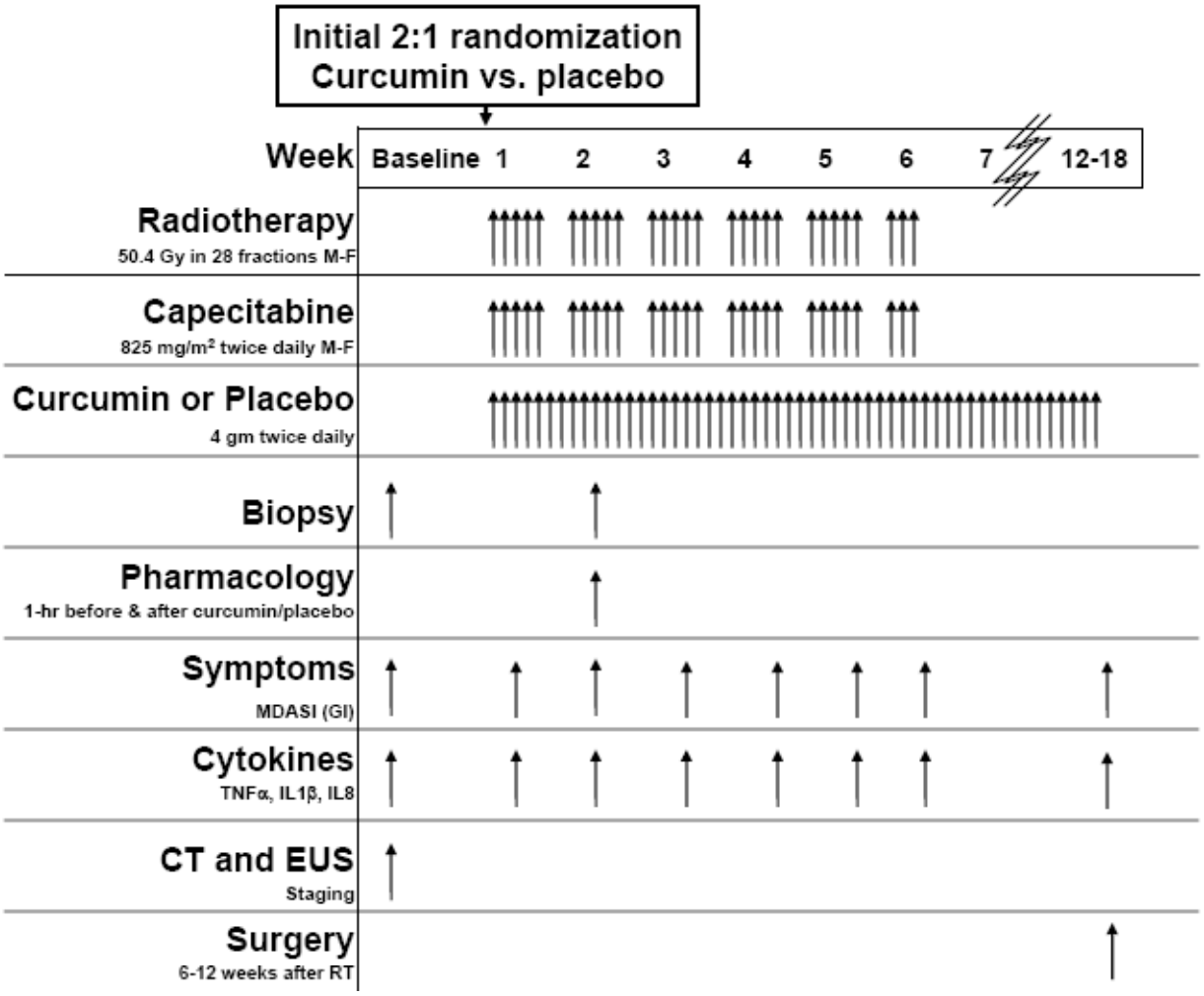
4.2.10 Sorivudine and brivudine use within 4 weeks of the start of study treatment.

4.3 Prohibited Medications

Aluminum hydroxide- and magnesium hydroxide-containing antacids such as Maalox should not be used during this study because they increase the AUC and Cmax of capecitabine. Because clinically significant increases in prothrombin time (PT) and INR in patients who were stabilized on anticoagulants have been shown, patients on Coumadin® must be changed to Lovenox® at least 1 week prior to starting capecitabine. Low dose (1 mg) Coumadin® is allowed. Patients on phenytoin must have their phenytoin and albumin levels checked weekly during chemoradiation. Vaccination with live vaccines should not be instituted during treatment with capecitabine.

5.0 Treatment Plan

5.1 Schema



5.2 Radiation Therapy (Patients can elect to receive XRT treatments only at MDACC satellite radiation centers. However, all patients must return to MDACC for evaluations during chemoradiation as required by the protocol.)

5.2.1 Simulation will be done using a CT-simulator.

5.2.2 Equipment: Megavoltage photons will be used to deliver radiotherapy.

5.2.3 Target Volumes:

5.2.3.1 Gross tumor volume (GTV): The GTV will comprise the pre-operative tumor volume and any involved lymph nodes, as defined by pre-operative CT scans and/or colonoscopy.

5.2.3.2 Clinical target volume 1 (CTV1): The CTV for the initial course of radiotherapy will comprise the GTV, the perirectal nodes, the presacral nodes and the internal iliac nodes. The external iliac nodes may also be included, at the discretion of the treating radiation oncologist.

5.2.3.3 Clinical target volume 2 (CTV2): The CTV for the boost will comprise the GTV.

5.2.3.4 Typical field design: A three-field technique (posteroanterior, right and left lateral fields) will be used to administer pelvic radiotherapy (to CTV1) with the patient on a belly board. Pelvic fields will extend from the L5–S1 interspace to the bottom of the obturator foramen or 3 cm below the tumor, whichever was lower.Laterally the field will extend 2 cm lateral to

the bony pelvis at its widest point. The anterior border of the lateral fields will be individually shaped to encompass the common iliac and internal iliac nodes for all tumors and the external iliac nodes for T4 tumors invading the vagina, prostate, or urinary bladder. The posterior border will include the entire sacrum with a 1-cm margin posteriorly. Fields will initially be dosimetrically weighted 2 (posteroanterior portal): 1 (right lateral portal): 1 (left lateral portal), usually with 45° wedges on the lateral fields and adjusted to ensure adequate coverage and homogeneity. The boost will be delivered using opposed lateral fields targeting CTV2 with a margin of 2-3 cm circumferentially and with inclusion of the presacral space and sacrum in the field.

5.2.4 Dose: CTV1 will be treated to a dose of 45 Gy. This will be followed by a boost to CTV2 with a dose of 5.4 Gy. At the discretion of the treating radiation oncologist, up to an additional 3.6 Gy may be administered during the boost. All plans will be generated using 3D conformal techniques. At the discretion of the radiation oncologist, an intensity-modulated radiation therapy (IMRT) plan may be generated.

5.2.5 Fractionation: Radiotherapy will be delivered in 180 cGy once daily fractions for 28 fractions.

5.2.6 Dose constraints: Dose to the femoral heads will be limited to 50 Gy. The volume of small bowel getting > 45 Gy will be minimized. Dose heterogeneity will be less than 10%.

5.2.7 Radiotherapy will be interrupted for treatment related > Grade 2 non-hematological toxicity until it resolves to grade 1, then continued. If radiotherapy is interrupted, then capecitabine will also be interrupted. Please note that for any event which is apparent at baseline, the interrruption will apply according to the corresponding shift in toxicity grade if the investigator feels it is appropriate (e.g. if a patient has grade 1 asthenia at baseline which increases to grade 2 during treatment, this will be considered as a shift of 1 grade and treated as a grade 1 toxicity for radiation interruption purposes).

5.3 Chemotherapy

5.3.1 Capecitabine

Capecitabine will be given at 825 mg/m^2 PO BID only on days of radiation (Monday through Friday). Dose adjustment of capecitabine will be made as needed (see 5.3.1.3).

5.3.1.1 Cautions when used with other medications

5.3.1.1.1 Concurrent use of coumadin other than low dose (1 mg) coumadin used for line patency is contraindicated. Patients on coumadin must be changed to Lovenox at least 1 week prior to starting capecitabine.

5.3.1.1.2 Increased phenytoin plasma concentrations have been reported during concomitant use of Capecitabine with phenytoin, suggesting a potential interaction. Patients taking phenytoin concomitantly with Capecitabine should be monitored weekly for increased phenytoin plasma concentrations and associated clinical symptoms.

5.3.1.1.3 Allopurinol. Oxypurinol, a metabolite of allopurinol, can potentially interfere with 5-FU anabolism via orotate phosphoribosyltransferase. Although this was originally used as a strategy to protect normal tissues from 5-FU-associated toxicity, further laboratory studies suggested possible antagonism of the anticancer activity of 5-FU in some tumor models. If a patient is receiving allopurinol, the need for taking this medicine should be ascertained. If possible, allopurinol should be discontinued prior to starting on this regimen, and another agent substituted for it.

5.3.1.1.4 Cimetidine: Because cimetidine can decrease the clearance of 5-FU, patients should not enter on this study until the cimetidine is discontinued. Ranitidine or a drug from another anti-ulcer class can be substituted for cimetidine if necessary.

5.3.1.1.5 Sorivudine and Brivudine: A metabolite of the above two investigational antiviral agents, 5-bromovinyluracil, is a potent inhibitor of dihydropyrimidine dehydrogenase, the enzyme that catabolizes 5-FU. Patients should not receive concurrent therapy with either of these antiviral agents while receiving capecitabine. If a patient has received prior sorivudine or brivudine, then at least four weeks must elapse before the patient receives capecitabine therapy.

5.3.1.2 Dosing

Capecitabine should be given approximately 12 hours apart (morning and evening), and taken within 30 minutes after the ingestion of food with approximately 200 mls of water (not fruit juices).

5.3.1.3 Dose modification guidelines of Capecitabine

Dose Modifications: For any event which is apparent at baseline, the dose modifications will apply according to the corresponding shift in toxicity grade if the investigator feels it is appropriate (e.g. if a patient has grade 1 asthenia at baseline which increases to grade 2 during treatment, this will be considered as a shift of 1 grade and treated as a grade 1 toxicity for dose modification purposes). Capecitabine treatment interruptions are regarded as lost treatment days and missed doses should not be replaced; the planned treatment schedule should be maintained. Once a dose has been reduced it should not be increased at a later time.
Toxicity **NCI Grade *(a)* (Value)**
Neutropenia Capecitabine will be held for >grade 2 neutropenia. Capecitabine can be restarted at 75% of the dose after recovery of neutropenia to grade 1. Patients will be removed from the study if they do not recover from the neutropenia.
Neutropenic fever (grade 4 neutropenia and grade 2 fever) Interrupt capecitabine until resolved to grade 0, then continue at 75% of starting dose. Patients will be removed from the study if they do not

recover from the neutropenic fever.
Other hematological toxicities No dose reductions or interruptions will be required for anemia as it can be satisfactorily managed by transfusions, if necessary.
Diarrhea [all; stools/day > pretreatment] 1 (2-3 stools/day) : Maintain dose 2 (4-6 stools/day)Interrupt until grade 0-1, continue according to number of appearances 1st : 75% of starting dose. 2nd : 50% of starting dose. 3rd : 50% of starting dose. 3 (7-9 stools/day)Interrupt until grade 0-1, continue according to number of appearances 1st : 75% of starting dose. 2nd : 50% of starting dose. 3rd : 50% of starting dose. 4 (10 stools/day)Interrupt until grade 0-1, then continue at 50% of starting dose.
Hand-Foot Syndrome 1 Maintain dose 2 Interrupt until grade 0-1, then continue at 75% of starting dose. 3 Interrupt until grade 0-1, then continue at 50% of starting dose. 4 (not applicable)
Other non-hematological toxicity For toxicities which are considered by the Investigator unlikely to develop into serious or life–threatening events and which do not result in a delay or interruption of therapy (e.g. alopecia, altered taste etc.), treatment will be continued at the same dose without reduction or interruption. 1 Maintain dose 2 Interrupt until grade 0-1, continue according to number of appearances of same toxicity 1st : 75% of starting dose. 2nd : 50% of starting dose. 3rd : 50% of starting dose. 3 Interrupt until grade 0-1, continue according to number of appearances of same toxicity 1st : 75% of starting dose. 2nd : 50% of starting dose. 3rd : 50% of starting dose. 4 Interrupt until grade 0-1, then continue at 50% of starting dose.
a. National Cancer Institute Common Toxicity Criteria Version 3.0, see Appendix B.

5.3.1.4 Special Instructions Regarding Treatment of Toxicity;

5.3.1.4.1 Grade 2/3 Hand–Foot Skin Reaction
Treat symptomatically (recommended use of emollients).
The use of vitamin B6 Pyridoxine has been reported to be of possible benefit and is permitted for symptomatic or secondary prophylactic treatment of hand-foot skin reaction.

5.3.1.4.2 Neutropenia

When the ANC has recovered to 1,500/mm^3 and fever or infection has resolved, the patient may restart treatment, with capecitabine at 75% of the starting dose if the patient had > grade 2 neutropenia, and at 100% of the starting dose if the patient did not have > grade 2 neutropenia.

5.3.1.4.3 Diarrhea

A three-step plan to manage diarrhea will be used. The goal will be to keep the frequency of bowel movements to less than four per day. Patients will initially take Lomotil as needed. When that is no longer sufficient to control the increased frequency of bowel movement, patients take 2 lomotil every 3-4 hours, which is step 2. Subsequently, Imodium is added and alternated with Lomotil, which is step 3; 2 tablets of one or the other is taken every 2-3 hours. Delayed and immediate release narcotics will be used at the discretion of the treating physician. Infectious diarrhea must be considered as an etiology, particularly if diarrhea occurs during the first two weeks of radiation. Outpatient intravenous rehydration will be given in patients who become dehydrated. At the discretion of the treating physician, empiric use of a fluoroquinolone may be considered.

5.3.1.5 Warnings and Precautions

5.3.1.5.1 Renal Insufficiency: Patients with moderate renal impairment at baseline require dose reduction (See section 3.2.9.2, Dosage Regimen and Administration.) Patients with mild and moderate renal impairment at baseline should be carefully monitored for adverse events. Prompt interruption of therapy with subsequent dose adjustments will be made if a patient develops a grade 2 to 4 adverse event. patients will be taken off study if they do not recover. Capecitabine is contraindicated in patients with a creatinine clearance of < 30 ml/min.

5.3.1.5.2 Pregnancy/Nursing: Capecitabine may cause fetal harm when given to a pregnant woman. If the drug is used during pregnancy, or if the patient becomes pregnant while receiving this drug, the patient should be apprised of the potential hazard to the fetus. Women of childbearing potential should be advised to avoid becoming pregnant while receiving treatment with capecitabine. Because of the potential for serious adverse reactions in nursing infants from capecitabine, it is recommended that nursing be discontinued when receiving capecitabine therapy.

5.3.1.5.3 Coagulopathy: Patients receiving concomitant capecitabine and oral coumarin-derivative anticoagulant therapy should have their anticoagulant response (INR or prothrombin time) monitored frequently in order to adjust the anticoagulant dose accordingly. A clinically important Capecitabine-Warfarin drug interaction was demonstrated in a clinical pharmacology trial. Altered coagulation parameters and/or bleeding, including death, have been reported in patients taking capecitabine concomitantly with coumarin-derivative

anticoagulants such as warfarin and phenprocoumon. Postmarketing reports have shown clinically significant increases in prothrombin time (PT) and INR in patients who were stabilized on anticoagulants at the time capecitabine was introduced. These events occurred within several days and up to several months after initiating capecitabine therapy and, in a few cases, within one month after stopping capecitabine. These events occurred in patients with and without liver metastases. Age greater than 60 and a diagnosis of cancer independently predispose patients to an increased risk of coagulopathy.

5.3.1.5.4 Cardiotoxicity: The cardiotoxicity observed with capecitabine includes myocardial infarction/ischemia, angina, dysrhythmias, cardiac arrest, cardiac failure, sudden death, electrocardiographic changes, and cardiomyopathy. These adverse events may be more common in patients with a prior history of coronary artery disease.

5.3.1.5.5 General: This treatment is foreseen as a self-administered out-patient treatment, and in certain circumstances adverse events that could occur, such as diarrhea, or hand-foot syndrome can rapidly become serious. In the case where a patient experiences any toxicity between scheduled visits, the patient will be instructed to contact the clinic as soon as possible, for further directions, discontinuation of study medication, and/or treatment.

5.4 Curcumin or Placebo

Curcumin or placebo capsules will be given at 4 gm PO twice daily on an empty stomach throughout the chemoradiation therapy period and for six additional weeks. On days of radiation therapy, the capsules will be consumed approximately 1 hour before radiation therapy. On the day that blood is drawn for pharmacology of curcumin or placebo, the 1 hour interval between capsules and radiation may be prolonged (see 7.5). Whenever capecitabine is discontinued, curcumin or placebo will be discontinued as well.

5.5 Patient Monitoring and Supportive Care

Patients will be evaluated weekly by clinicians participating in their care. The clinician will be responsible for performing a history, physical exam, and monitoring lab work. The common acute effects of chemoradiation are managed with aggressive outpatient supportive care. The judicious use of prophylactic antiemetics and a three-step plan to manage diarrhea are used. The goal is to keep the frequency of bowel movements to less than four per day. Patients are initially instructed to take Lomotil as needed. When that is no longer sufficient to control the increased frequency of bowel movement, patients take 2 lomotil every 3-4 hours, which is step 2. Subsequently, Imodium is added and alternated with Lomotil, which is step 3; 2 tablets of one or the other is taken every 2-3 hours. Delayed and immediate release narcotics are used regularly. Another common acute reaction is desquamation of the perineal skin and genitalia, which usually occurs with irradiation of low rectal lesions. These reactions can be effectively managed with a lanolin-containing barrier cream used in the perianal area, anusol suppositories for anal canal pain, and Aquaphor for the anterior skin reactions. Moist desquamation often requires narcotic pain medication, sitz baths, and the use of a hydrogel dressing (Vigilon

or Cool Magic).

5.6 Surgery and Adjuvant Therapy

Patients will be evaluated by a surgeon 6-12 weeks after completion of radiotherapy for consideration of surgery. A separate consent form will be obtained for surgery. Further systemic therapy may be offered at the discretion of the treating oncologist.

6.0 Pretreatment evaluation

6.1 Prior to Study Enrollment:

6.1.1 Patients must have the following tests before study enrollment:

6.1.1.1 Blood chemistries (bilirubin, alkaline phosphatase, ALT, AST, BUN, creatinine, phosphorus, calcium, glucose, total protein, albumin, and electrolytes [sodium, potassium, carbon dioxide, and chloride], Complete blood count (CBC: hemoglobin, hematocrit, WBC with differential blood cell counts (neutrophils, bands, lymphocytes, monocytes, eosinophils, basophils) and platelets), carcinoembryonic antigen (CEA), urinalysis, all within 10 days of enrollment. PT and INR will be done if patient is on coumadin and phenytoin levels will be checked in patients on phenytoin.

6.1.1.2 Urine pregnancy test within 72 hours of enrollment in women of child-bearing potential

6.1.1.3 Radiographic studies (CXR or chest CT; CT of the abdomen and pelvis) within 30 days of enrollment

6.1.1.4 Colonoscopy within 3 months of enrollment and flexible sigmoidoscopy/ EUS within 30 days of enrollment

6.1.1.5 Endoscopic biopsy prior to study enrollment

6.1.2 After consent but prior to initiating chemoradiation therapy, additional tests for research purposes include:

6.1.2.1 MD Anderson symptom inventory (MDASI) questionnaire gastrointestinal module (appendix F)

6.1.2.2 Brief Fatigue Inventory (BFI) (appendix E)

6.1.2.3 Blood collection for cytokine analysis

7.0 Evaluation During Study

During chemoradiation therapy, the patients will be assessed as follows:

7.1 Blood chemistries (bilirubin, alkaline phosphatase, ALT, AST, BUN, creatinine, phosphorus, calcium, glucose, total protein, albumin, and electrolytes [sodium, potassium, carbon dioxide, and chloride], and complete blood count (CBC: hemoglobin, hematocrit, WBC with differential blood cell counts (neutrophils, bands, lymphocytes, monocytes, eosinophils, basophils) and platelets) every week. Phenytoin levels will be checked in patients on phenytoin.

7.2 Weekly port-films

7.3 Weekly MDASI-GI and BFI questionnaires

7.4 Weekly blood collection for cytokine analysis

7.5 During week 2 (after at least 5 fractions of radiation therapy) of chemoradiation therapy:

a) Optional endoscopic biopsy

b) Optional blood collection for pharmacology (1 hour before and 1 hour after intake of curcumin or placebo)

7.6 Patients will be assessed by physician and research nurse at least once every 5 radiation fractions or as needed +/- 1 or 2 radiation fractions

Six-twelve weeks after chemoradiation therapy, patients will be restaged as follows:

7.9 Blood chemistries (bilirubin, alkaline phosphatase, ALT, AST, BUN, creatinine, phosphorus, calcium, glucose, total protein, albumin, and electrolytes [sodium, potassium, carbon dioxide, and chloride], complete blood count (CBC: hemoglobin, hematocrit, WBC with differential blood cell counts (neutrophils, bands, lymphocytes, monocytes, eosinophils, basophils and platelets), and CEA.

7.10 MDASI-GI and BFI questionnaires

7.11 Blood collection for cytokine analysis

7.12 Study Calendar:

Tests & Procedures	*Pre-treatment*	*During chemo-radiation*	*Week #2 of chemo-radiation*	*6-12 weeks after chemo-radiation (restaging)*	*Follow-up (5)*
H&P; KPS	x (1)	weekly		x	
CBC (see 6.1)	x (1)	weekly		x	
Serum chemistries (see 6.1)	x (1)	weekly		x	
CEA	x (1)			x	
Phenytoin level (patients on phenytoin)	x (1)	weekly			
PT, INR (patients on coumadin)	x (1)	weekly		x	
Urinalysis	x (1)				
Urine pregnancy test (see 6.1)	x (2)				
Abd-Pelvic CT	x(3)				
Flexible sigmoidoscopy/EUS	x (3)				
Endoscopic ultrasound biopsy	x		x (6)		
CXR	x (3)				
Colonoscopy	x (4)				
MDASI, BFI	x	weekly	x	x	
Cytokines (see 9.2) (6)	x	weekly	x	x	
Pharmacology (see 9.1) (6)			x		
Telephone call (toxicity check)					x

(1) Within 15 days of enrollment
(2) Urine pregnancy test within 72 hours of enrollment, women of child bearing age only
(3) Within 30 days of enrollment
(4) Within 3 months of enrollment

(5) Telephone call 1 month +/- 1 week after last dose of curcumin/placebo.
(6) Optional

8.0 Evaluation After Treatment

After finishing chemoradiation treatments and "6 week maintenance (curcumin vs. placebo)", the patient will have a telephone follow-up visit 1 month +/- 1 week. Since most surgeries take place within the 1-3 weeks after the 6 week maintenance, the patient will typically be in surgical planning/ implementation/recovery. Furthermore, since most patients are from out of town, this follow-up visit is best accomplished via a telephone call. Therefore, toxicity check will be verbal and a note will be recorded in the patient's chart. Surgical toxicity will not be recorded since it is likely that patients will complain of incisional discomfort, abdominal pain, and symptoms related to surgery. Most side effects of chemoradiation would have resolved by the time of this telephone call.

Patients may receive adjuvant chemotherapy after surgery, at the discretion of the treating medical oncologist.

Standard-of-care follow-up evaluations will be undertaken per the directions of the treating medical oncologist. The patient chart will be reviewed periodically to capture failure and survival information.

9.0 Correlative Studies

9.1 Clinical pharmacology studies (optional)

9.1.1 Blood samples (20 ml) will be collected in lavender top tubes at the Endoscopy suite during the second week of CRT and processed as follows. This will be performed pre-dose and 1-hour +/- 10 minutes post-dose of curcumin or placebo. A third of the sample will be centrifuged immediately (1500 x g for 15 min at 4°C). The plasma is removed and stored at -20°C. The samples will be shipped in the frozen state for storage until ready for analysis in the core pharmacology lab. The remaining two-thirds of the sample will be transferred into a 50 cc conical tube to which is added an equal volume of PBS. The tube is inverted 10 times. The diluted blood is transferred to another 50 ml conical tube that contains an equal volume of Ficoll-Paque using a transfer pipette. The diluted blood is carefully laid over the Ficoll-Paque at room temperature. The tube is capped and centrifuged (700 x g for 30 min) at room temperature using a swinging bucket rotor. Cells (peripheral blood mononuclear cells, PBMC) are obtained at the resulting interface, transferred to another tube and washed once with PBS. After centrifugation, cells are then resuspended, and samples saved for immunocytochemical analyses.

9.1.1.1 Plasma curcumin, curcumin glucuronide, curcumin sulfate, demethoxycurcumin,and bisdemethoxycurcumin will be measured using high-performance liquid chromatography and mass spectrometry.

9.1.1.2 Curcumin and its metabolites will be expressed in micrograms of drug per milliliter of plasma.
9.1.1.3 Results will be expressed as means +/- standard deviations.
9.1.1.4 PBMC will be used for immunocytochemical analysis.

9.1.2 Biopsies (tumor) will be obtained during the second week of chemoradiation therapy and at restaging. Surgical specimen will also be collected.

9.1.2.1 Biopsy tissue and surgical tissue will be immediately frozen in liquid nitrogen and stored at -80°C. The tissue will be homogenized (liquid nitrogen cooled mortar and pestle, cold spatula transfer to cooled eppendorf, cryocentrifuged, digested, homogenized in ultrasonic homogenizer) and then analyzed for targets (curcumin and its metabolites) as noted above.
9.1.2.2 Curcumin and its metabolites will be expressed in micrograms of drug per milligram of wet tissue weight.
9.1.1.3 Results will be expressed as means +/- standard deviations.

9.2 Cytokine assays (optional)

These will be performed by personnel who are blinded to the treatment status of the patients.

9.2.1 Blood will be collected in lavendar top tubes at study initiation and weekly during chemoradiation for cytokine analyses. This will be stored in the laboratory until bunched samples can be analyzed in the core cytokine lab. The cytokines assayed will include IL-1b, IL-8 and TNF-a.

9.3 Surrogate marker analyses

These will be performed by personnel who are blinded to the treatment status of the patients.

Fresh frozen tumor from biopsy and surgical specimens will be processed for correlative studies as noted below:

9.3.1 A portion of tumor tissue will be snap frozen for measurement of drug levels as noted in 9.1.2.
9.3.2 A portion will be frozen for Western blot analysis, electrophoretic mobility shift assay, and microarray analysis. Tissue will be homogenized and probed with specific antibodies for Western blot analysis of key proteins including bcl-2, bax, PARP, VEGF, cyclin D1, and cox-2. Nuclear extracts will be prepared for EMSA of NF-kB DNA-binding.
9.3.3 Another portion will be processed for immunohistochemical analysis including p65, p-stat3, cox-2, VEGF, Ki-67, and CD31 evaluation.
9.3.4 The remainder will be frozen for further analysis.
9.3.5 Labeled sterile Nunc tubes will be used to store tissue samples.
9.3.6 All tissue not mentioned above will be routinely processed as per the pathology department.

10.0 Criteria for Toxicity

Patients will be evaluated each week during chemoradiation just before surgery and 1 month +/-

2 days after surgery by a physician and research nurse regarding the occurrence and nature of any adverse events. Appendix B provides the grades to be assigned for various types of toxicities.

10.1 Off-study criteria
Patients who have documented clinical progression of disease that is discovered upon symptomatic progression during therapy will be taken off study. Patients will be taken off study if they fail to complete protocol therapy for reasons other than treatment related toxicity or tumor progression and will not be counted in the analysis. Patients will be taken off study if they miss two consecutive appointments.

10.2 Criteria for Discontinuing Therapy
Treatment will be discontinued if an unpredictable, irreversible toxicity develops, or patient chooses to discontinue treatment/study.

10.3 See 12.0 for reporting requirements.

11.0 Data and Protocol Management

11.1 Patients will be registered by calling a Radiation Oncology Research Nurse, who will then register them on the MDACC computerized Clinical Oncology Research System (CORe).

12.0 Reporting Guidelines

12.1 All serious adverse events described in Appendix A will be reported to the IRB. Serious adverse events will be delivered to the Office of Research Education and Regulatory Management - IND office and will be submitted to the FDA by the Project Manager for IND safety, according to 21 CFR 312.32.
12.2 Surgical complications will not be reported.
12.3 Lymphopenia, high blood sugar and electrolyte abnormalities will not be reported.
12.4 Toxicities grade 2 and above will be captured in PDMS data.

13.0 Statistical Considerations

The Department of Biostatistics will create a randomization website to be used by the pharmacists to assign treatments to patients. Patients will be randomized to control and experimental treatment arms in a 1:2 ratio. That is, the probability of a patient being randomized to the control arm (placebo) is 1/3, while the probability of a patient being randomized to the experimental arm (curcumin) is 2/3. The primary outcome for this trial is the pCR rate, which is evaluated at the time of surgery. We expect the pCR rate to be 18% in the control group (3, 5, 15).

Our target pCR rate is 36% or more in the curcumin treatment group. For those patients randomized to curcumin we will employ a two-stage design with a significance level of 10% and 80% power. We will treat 16 patients in the first stage, and if there are 3 or more patients with pCR among these first 16 patients, we will treat 14 additional patients in the second stage, for a total of 30 patients. Curcumin will be considered for further development if there are 9 or more patients with pCR among these 30 patients. The probability of stopping the study after the first stage if the pCR rate is 18% is 0.43, while the probability of stopping the study after the first

stage if the pCR rate is 36% is 0.04. The expected sample size for this two-stage design is 24 patients, and the actual significance level is 7.5%." and replace it with "The expected sample size if the pCR rate is 18% is 24, and the actual significance level is 7.5%. We will randomize 15 patients to the placebo arm, yielding a total maximum sample size of 45 patients. We will accrue patients at a rate of 4 patients per month.

Secondary Outcomes

Downstaging

We will also compare the 2 treatment arms with respect to the proportions of patients that are downstaged at the time of surgery. We expect the proportion of patients in the control arm that will be downstaged is 60% (3, 5, 15). With 15 patients in the control arm and 30 patients in the experimental arm, we will have 82% power to detect an increase in the proportion of patients that are downstaged to 95%.

Correlative Studies

For the experimental treatment we will compare the concentration of curcumin in the tumor with the curcumin concentration levels in serum. We will find the difference in curcumin concentration levels in tumor and serum for each patient (tumor – serum), and then we will use a paired t-test to determine whether the mean difference is statistically significant.

For the experimental treatment we will estimate the correlation between the curcumin level in tumor tissue and 1) NF-kB activity, and 2) pCR. We will also estimate the correlation between the curcumin level in serum and these same variables.

We will use a logistic regression model with pCR as the dependent variable to assess the association between pCR and NF-kB activity and treatment.

We will also compare between treatment groups changes in selected components of known relevant pathways in both normal and tumor tissue. These include bcl-2, bax, PARP, VEGF, cyclin D1, p-stat3, Ki-67, CD31and cox-2.

Quality of Life and Toxicity

Patients will complete validated quality of life (QoL) instruments at baseline and weekly during chemoradiation. These instruments include the MD Anderson Symptom Inventory-Gastrointestinal (MDASI-GI) and Brief Fatigue Inventory (BFI). A panel of cytokines (IL-1b, IL-8 and TNF-a) will be assayed at these same evaluation times. We will estimate the correlation between the various cytokine levels and the various QoL instrument scores.

We will also compare changes from baseline in cytokine levels between treatment groups using a repeated measures analysis of variance model. We will construct side-by-side boxplots of changes from baseline in cytokine levels by treatment group for each evaluation time. We will also construct side-by-side boxplots of changes from baseline of QoL instrument scores by treatment group for each evaluation time.

14.0 References

1. Jemal A, Siegel R, Ward E*, et al.* Cancer statistics, 2006. *CA Cancer J Clin* 2006;56:106-130.
2. Sauer R, Becker H, Hohenberger W*, et al.* Preoperative versus postoperative chemoradiotherapy for rectal cancer. *N Engl J Med* 2004;351:1731-1740.
3. Das P, Skibber JM, Rodriguez-Bigas MA*, et al.* Clinical and pathologic predictors of locoregional recurrence, distant metastasis, and overall survival in patients treated with chemoradiation and mesorectal excision for rectal cancer. *Am J Clin Oncol* 2006;29:219-224.
4. Janjan NA, Crane CN, Feig BW*, et al.* Prospective trial of preoperative concomitant boost radiotherapy with continuous infusion 5-fluorouracil for locally advanced rectal cancer. *Int J Radiat Oncol Biol Phys* 2000;47:713-718.
5. Das P, Skibber JM, Rodriguez-Bigas MA*, et al.* Predictors of tumor response and downstaging in patients treated with preoperative chemoradiation for rectal cancer. *Proceedings of the American Society for Therapeutic Radiology and Oncology 2006 (abstract)* 2006.
6. Janjan NA, Crane C, Feig BW*, et al.* Improved overall survival among responders to preoperative chemoradiation for locally advanced rectal cancer. *Am J Clin Oncol* 2001;24:107-112.
7. Janjan NA, Abbruzzese J, Pazdur R*, et al.* Prognostic implications of response to preoperative infusional chemoradiation in locally advanced rectal cancer. *Radiother Oncol* 1999;51:153-160.
8. Berger C, de Muret A, Garaud P*, et al.* Preoperative radiotherapy (RT) for rectal cancer: predictive factors of tumor downstaging and residual tumor cell density (RTCD): prognostic implications. *Int J Radiat Oncol Biol Phys* 1997;37:619-627.
9. Kaminsky-Forrett MC, Conroy T, Luporsi E*, et al.* Prognostic implications of downstaging following preoperative radiation therapy for operable T3-T4 rectal cancer. *Int J Radiat Oncol Biol Phys* 1998;42:935-941.
10. Valentini V, Coco C, Picciocchi A*, et al.* Does downstaging predict improved outcome after preoperative chemoradiation for extraperitoneal locally advanced rectal cancer? A long-term analysis of 165 patients. *Int J Radiat Oncol Biol Phys* 2002;53:664-674.
11. Crane CH, Skibber JM, Feig BW*, et al.* Response to preoperative chemoradiation increases the use of sphincter-preserving surgery in patients with locally advanced low rectal carcinoma. *Cancer* 2003;97:517-524.
12. Janjan NA, Khoo VS, Abbruzzese J*, et al.* Tumor downstaging and sphincter preservation with preoperative chemoradiation in locally advanced rectal cancer: the M. D. Anderson Cancer Center experience. *Int J Radiat Oncol Biol Phys* 1999;44:1027-1038.
13. Bonnen M, Crane C, Vauthey JN*, et al.* Long-term results using local excision after preoperative chemoradiation among selected T3 rectal cancer patients. *Int J Radiat Oncol Biol Phys* 2004;60:1098-1105.
14. Fu KK, Pajak TF, Trotti A*, et al.* A Radiation Therapy Oncology Group (RTOG) phase III randomized study to compare hyperfractionation and two variants of accelerated fractionation to standard fractionation radiotherapy for head and neck squamous cell carcinomas: first report of RTOG 9003. *Int J Radiat Oncol Biol Phys* 2000;48:7-16.
15. Krishnan S, Janjan NA, Skibber J, et al. Phase II study of capecitabine (Xeloda®) and concomitant boost radiotherapy in patients with locally advanced rectal cancer. Int J Radiat Oncol Biol Phys 2006 66:762-771
16. Czito BG, Willett CG, Bendell JC*, et al.* Increased toxicity with gefitinib, capecitabine, and radiation therapy in pancreatic and rectal cancer: phase I trial results. *J Clin Oncol* 2006;24:656-662.
17. Mohiuddin M, Winter K, Mitchell E*, et al.* Randomized phase II study of neoadjuvant

combined-modality chemoradiation for distal rectal cancer: Radiation Therapy Oncology Group Trial 0012. *J Clin Oncol* 2006;24:650-655.

18. Ryan DP, Niedzwiecki D, Hollis D*, et al.* Phase I/II study of preoperative oxaliplatin, fluorouracil, and external-beam radiation therapy in patients with locally advanced rectal cancer: Cancer and Leukemia Group B 89901. *J Clin Oncol* 2006;24:2557-2562.
19. McMullen KP, Blackstock AW. Chemoradiation with novel agents for rectal cancer. *Clin Colorectal Cancer* 2002;2:24-30.
20. Zhu AX, Willett CG. Chemotherapeutic and biologic agents as radiosensitizers in rectal cancer. *Semin Radiat Oncol* 2003;13:454-468.
21. Chakravarti A, Loeffler JS, Dyson NJ. Insulin-like growth factor receptor I mediates resistance to anti-epidermal growth factor receptor therapy in primary human glioblastoma cells through continued activation of phosphoinositide 3-kinase signaling. *Cancer Res* 2002;62:200-207.
22. Arteaga CL. EGF receptor as a therapeutic target: patient selection and mechanisms of resistance to receptor-targeted drugs. *J Clin Oncol* 2003;21:289s-291s.
23. Sharma RA, Gescher AJ, Steward WP. Curcumin: the story so far. *Eur J Cancer* 2005;41:1955-1968.
24. Ammon HP, Wahl MA. Pharmacology of Curcuma longa. *Planta Med* 1991;57:1-7.
25. Aggarwal BB, Kumar A, Bharti AC. Anticancer potential of curcumin: preclinical and clinical studies. *Anticancer Res* 2003;23:363-398.
26. Aggarwal BB, Shishodia S. Molecular targets of dietary agents for prevention and therapy of cancer. *Biochem Pharmacol* 2006;71:1397-1421.
27. Tomita M, Kawakami H, Uchihara JN*, et al.* Curcumin (diferuloylmethane) inhibits constitutive active NF-kappaB, leading to suppression of cell growth of human T-cell leukemia virus type I-infected T-cell lines and primary adult T-cell leukemia cells. *Int J Cancer* 2006;118:765-772.
28. Shishodia S, Amin HM, Lai R*, et al.* Curcumin (diferuloylmethane) inhibits constitutive NF-kappaB activation, induces G1/S arrest, suppresses proliferation, and induces apoptosis in mantle cell lymphoma. *Biochem Pharmacol* 2005;70:700-713.
29. Siwak DR, Shishodia S, Aggarwal BB*, et al.* Curcumin-induced antiproliferative and proapoptotic effects in melanoma cells are associated with suppression of IkappaB kinase and nuclear factor kappaB activity and are independent of the B-Raf/mitogen-activated/extracellular signal-regulated protein kinase pathway and the Akt pathway. *Cancer* 2005;104:879-890.
30. Li L, Aggarwal BB, Shishodia S*, et al.* Nuclear factor-kappaB and IkappaB kinase are constitutively active in human pancreatic cells, and their down-regulation by curcumin (diferuloylmethane) is associated with the suppression of proliferation and the induction of apoptosis. *Cancer* 2004;101:2351-2362.
31. Wei SC, Lin YS, Tsao PN*, et al.* Comparison of the anti-proliferation and apoptosis-induction activities of sulindac, celecoxib, curcumin, and nifedipine in mismatch repair-deficient cell lines. *J Formos Med Assoc* 2004;103:599-606.
32. Bharti AC, Shishodia S, Reuben JM*, et al.* Nuclear factor-kappaB and STAT3 are constitutively active in CD138+ cells derived from multiple myeloma patients, and suppression of these transcription factors leads to apoptosis. *Blood* 2004;103:3175-3184.
33. Chan MM, Fong D, Soprano KJ*, et al.* Inhibition of growth and sensitization to cisplatin-mediated killing of ovarian cancer cells by polyphenolic chemopreventive agents. *J Cell Physiol* 2003;194:63-70.
34. Mehta K, Pantazis P, McQueen T*, et al.* Antiproliferative effect of curcumin (diferuloylmethane) against human breast tumor cell lines. *Anticancer Drugs* 1997;8:470-481.

35. Furness MS, Robinson TP, Ehlers T*, et al.* Antiangiogenic agents: studies on fumagillin and curcumin analogs. *Curr Pharm Des* 2005;11:357-373.
36. Zheng M, Ekmekcioglu S, Walch ET*, et al.* Inhibition of nuclear factor-kappaB and nitric oxide by curcumin induces G2/M cell cycle arrest and apoptosis in human melanoma cells. *Melanoma Res* 2004;14:165-171.
37. Moragoda L, Jaszewski R, Majumdar AP. Curcumin induced modulation of cell cycle and apoptosis in gastric and colon cancer cells. *Anticancer Res* 2001;21:873-878.
38. Aggarwal BB, Takada Y. Pro-apototic and anti-apoptotic effects of tumor necrosis factor in tumor cells. Role of nuclear transcription factor NF-kappaB. *Cancer Treat Res* 2005;126:103-127.
39. Ondrey FG, Dong G, Sunwoo J*, et al.* Constitutive activation of transcription factors NF-(kappa)B, AP-1, and NF-IL6 in human head and neck squamous cell carcinoma cell lines that express pro-inflammatory and pro-angiogenic cytokines. *Mol Carcinog* 1999;26:119-129.
40. Aggarwal BB. Nuclear factor-kappaB: the enemy within. *Cancer Cell* 2004;6:203-208.
41. Shishodia S, Aggarwal BB. Nuclear factor-kappaB activation mediates cellular transformation, proliferation, invasion angiogenesis and metastasis of cancer. *Cancer Treat Res* 2004;119:139-173.
42. Karin M. Nuclear factor-kappaB in cancer development and progression. *Nature* 2006;441:431-436.
43. Magne N, Toillon RA, Bottero V*, et al.* NF-kappaB modulation and ionizing radiation: mechanisms and future directions for cancer treatment. *Cancer Lett* 2006;231:158-168.
44. Beg AA, Baltimore D. An essential role for NF-kappaB in preventing TNF-alpha-induced cell death. *Science* 1996;274:782-784.
45. Garg A, Aggarwal BB. Nuclear transcription factor-kappaB as a target for cancer drug development. *Leukemia* 2002;16:1053-1068.
46. Bharti AC, Aggarwal BB. Nuclear factor-kappa B and cancer: its role in prevention and therapy. *Biochem Pharmacol* 2002;64:883-888.
47. Luo JL, Kamata H, Karin M. IKK/NF-kappaB signaling: balancing life and death--a new approach to cancer therapy. *J Clin Invest* 2005;115:2625-2632.
48. Karin M, Greten FR. NF-kappaB: linking inflammation and immunity to cancer development and progression. *Nat Rev Immunol* 2005;5:749-759.
49. Garg AK, Buchholz TA, Aggarwal BB. Chemosensitization and radiosensitization of tumors by plant polyphenols. *Antioxid Redox Signal* 2005;7:1630-1647.
50. Aggarwal S, Ichikawa H, Takada Y*, et al.* Curcumin (diferuloylmethane) down-regulates expression of cell proliferation and antiapoptotic and metastatic gene products through suppression of IkappaBalpha kinase and Akt activation. *Mol Pharmacol* 2006;69:195-206.
51. Aggarwal S, Takada Y, Singh S*, et al.* Inhibition of growth and survival of human head and neck squamous cell carcinoma cells by curcumin via modulation of nuclear factor-kappaB signaling. *Int J Cancer* 2004;111:679-692.
52. Singh S, Aggarwal BB. Activation of transcription factor NF-kappa B is suppressed by curcumin (diferuloylmethane) [corrected]. *J Biol Chem* 1995;270:24995-25000.
53. Jobin C, Bradham CA, Russo MP*, et al.* Curcumin blocks cytokine-mediated NF-kappa B activation and proinflammatory gene expression by inhibiting inhibitory factor I-kappa B kinase activity. *J Immunol* 1999;163:3474-3483.
54. Plummer SM, Holloway KA, Manson MM*, et al.* Inhibition of cyclo-oxygenase 2 expression in colon cells by the chemopreventive agent curcumin involves inhibition of NF-kappaB activation via the NIK/IKK signalling complex. *Oncogene* 1999;18:6013-6020.
55. Su CC, Chen GW, Lin JG*, et al.* Curcumin inhibits cell migration of human colon cancer

colo 205 cells through the inhibition of nuclear factor kappa B /p65 and down-regulates cyclooxygenase-2 and matrix metalloproteinase-2 expressions. *Anticancer Res* 2006;26:1281-1288.

56. Lind DS, Hochwald SN, Malaty J*, et al.* Nuclear factor-kappa B is upregulated in colorectal cancer. *Surgery* 2001;130:363-369.
57. Bours V, Dejardin E, Goujon-Letawe F*, et al.* The NF-kappa B transcription factor and cancer: high expression of NF-kappa B- and I kappa B-related proteins in tumor cell lines. *Biochem Pharmacol* 1994;47:145-149.
58. Singh S, Khar A. Biological effects of curcumin and its role in cancer chemoprevention and therapy. *Anticancer Agents Med Chem* 2006;6:259-270.
59. Goel A, Boland CR, Chauhan DP. Specific inhibition of cyclooxygenase-2 (COX-2) expression by dietary curcumin in HT-29 human colon cancer cells. *Cancer Lett* 2001;172:111-118.
60. Zhang F, Altorki NK, Mestre JR*, et al.* Curcumin inhibits cyclooxygenase-2 transcription in bile acid- and phorbol ester-treated human gastrointestinal epithelial cells. *Carcinogenesis* 1999;20:445-451.
61. Berger AL, Randak CO, Ostedgaard LS*, et al.* Curcumin stimulates cystic fibrosis transmembrane conductance regulator Cl- channel activity. *J Biol Chem* 2005;280:5221-5226.
62. Korutla L, Kumar R. Inhibitory effect of curcumin on epidermal growth factor receptor kinase activity in A431 cells. *Biochim Biophys Acta* 1994;1224:597-600.
63. Collett GP, Campbell FC. Curcumin induces c-jun N-terminal kinase-dependent apoptosis in HCT116 human colon cancer cells. *Carcinogenesis* 2004;25:2183-2189.
64. Jeong WS, Kim IW, Hu R*, et al.* Modulation of AP-1 by natural chemopreventive compounds in human colon HT-29 cancer cell line. *Pharm Res* 2004;21:649-660.
65. Chen H, Zhang ZS, Zhang YL*, et al.* Curcumin inhibits cell proliferation by interfering with the cell cycle and inducing apoptosis in colon carcinoma cells. *Anticancer Res* 1999;19:3675-3680.
66. Bharti AC, Donato N, Aggarwal BB. Curcumin (diferuloylmethane) inhibits constitutive and IL-6-inducible STAT3 phosphorylation in human multiple myeloma cells. *J Immunol* 2003;171:3863-3871.
67. Mukhopadhyay A, Bueso-Ramos C, Chatterjee D*, et al.* Curcumin downregulates cell survival mechanisms in human prostate cancer cell lines. *Oncogene* 2001;20:7597-7609.
68. Chen A, Xu J. Activation of PPAR{gamma} by curcumin inhibits Moser cell growth and mediates suppression of gene expression of cyclin D1 and EGFR. *Am J Physiol Gastrointest Liver Physiol* 2005;288:G447-456.
69. Bhaumik S, Jyothi MD, Khar A. Differential modulation of nitric oxide production by curcumin in host macrophages and NK cells. *FEBS Lett* 2000;483:78-82.
70. Gao X, Kuo J, Jiang H*, et al.* Immunomodulatory activity of curcumin: suppression of lymphocyte proliferation, development of cell-mediated cytotoxicity, and cytokine production in vitro. *Biochem Pharmacol* 2004;68:51-61.
71. Jana NR, Dikshit P, Goswami A*, et al.* Inhibition of proteasomal function by curcumin induces apoptosis through mitochondrial pathway. *J Biol Chem* 2004;279:11680-11685.
72. Pal S, Choudhuri T, Chattopadhyay S*, et al.* Mechanisms of curcumin-induced apoptosis of Ehrlich's ascites carcinoma cells. *Biochem Biophys Res Commun* 2001;288:658-665.
73. Bush JA, Cheung KJ, Jr., Li G. Curcumin induces apoptosis in human melanoma cells through a Fas receptor/caspase-8 pathway independent of p53. *Exp Cell Res* 2001;271:305-314.
74. Khar A, Ali AM, Pardhasaradhi BV*, et al.* Antitumor activity of curcumin is mediated through the induction of apoptosis in AK-5 tumor cells. *FEBS Lett* 1999;445:165-168.

75. Krishnan S, Sandur SK, Shentu S, *et al.* Curcumin enhances colorectal cancer cell radiosensitivity by suppressing the radiation-induced nuclear factor-kappaB (NF-kB) pathway. *Int J Radiat Oncol Biol Phys (*66): S547 (abstract)
76. O'Connell MJ, Martenson JA, Wieand HS*, et al.* Improving adjuvant therapy for rectal cancer by combining protracted-infusion fluorouracil with radiation therapy after curative surgery. *N Engl J Med* 1994;331:502-507.
77. Twelves C, Wong A, Nowacki MP*, et al.* Capecitabine as adjuvant treatment for stage III colon cancer. *N Engl J Med* 2005;352:2696-2704.
78. Dunst J, Reese T, Sutter T*, et al.* Phase I trial evaluating the concurrent combination of radiotherapy and capecitabine in rectal cancer. *J Clin Oncol* 2002;20:3983-3991.
79. Van Cutsem E, Findlay M, Osterwalder B*, et al.* Capecitabine, an oral fluoropyrimidine carbamate with substantial activity in advanced colorectal cancer: results of a randomized phase II study. *J Clin Oncol* 2000;18:1337-1345.
80. Hoff PM, Ansari R, Batist G*, et al.* Comparison of oral capecitabine versus intravenous fluorouracil plus leucovorin as first-line treatment in 605 patients with metastatic colorectal cancer: results of a randomized phase III study. *J Clin Oncol* 2001;19:2282-2292.
81. Van Cutsem E, Twelves C, Cassidy J*, et al.* Oral capecitabine compared with intravenous fluorouracil plus leucovorin in patients with metastatic colorectal cancer: results of a large phase III study. *J Clin Oncol* 2001;19:4097-4106.
82. Kuttan R, Sudheeran PC, Josph CD. Turmeric and curcumin as topical agents in cancer therapy. *Tumori* 1987;73:29-31.
83. Cheng AL, Hsu CH, Lin JK*, et al.* Phase I clinical trial of curcumin, a chemopreventive agent, in patients with high-risk or pre-malignant lesions. *Anticancer Res* 2001;21:2895-2900.
84. Lao CD, Ruffin MTt, Normolle D*, et al.* Dose escalation of a curcuminoid formulation. *BMC Complement Altern Med* 2006;6:10.
85. Sharma RA, McLelland HR, Hill KA*, et al.* Pharmacodynamic and pharmacokinetic study of oral Curcuma extract in patients with colorectal cancer. *Clin Cancer Res* 2001;7:1894-1900.
86. Sharma RA, Euden SA, Platton SL*, et al.* Phase I clinical trial of oral curcumin: biomarkers of systemic activity and compliance. *Clin Cancer Res* 2004;10:6847-6854.
87. Garcea G, Berry DP, Jones DJ*, et al.* Consumption of the putative chemopreventive agent curcumin by cancer patients: assessment of curcumin levels in the colorectum and their pharmacodynamic consequences. *Cancer Epidemiol Biomarkers Prev* 2005;14:120-125.