Abbreviated Title: Chemo+antiPDL1+ vaccine in CRC

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Title: A Randomized Phase II trial of Standard of Care Alone or in Combination with Ad-CEA vaccine and Avelumab in Patients with Previously Untreated Metastatic or Unresectable Colorectal Cancer

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Investigational Agents:

Drug Name:	avelumab; anti-PD-L1 antibody; MSB0010718C	ETBX-011; adenoviral CEA vaccine
BB IND Number:	17056	17056
Sponsor:	Center for Cancer Research	Center for Cancer Research
Manufacturer:	EMD Serono, Inc.	Etubics Corporation

Commercial agents:

bevacizumab, 5-FU, leucovorin, oxaliplatin, and capecitabine will be purchased by the pharmacy at each participating institution

PRÉCIS

Background:

- Colorectal cancer (CRC) is the fourth most common cancer diagnosis in the United States and accounts for the second most cancer-related deaths.
- Programmed death ligand 1 (PD-L1) is a transmembrane protein that was first identified for its role in the maintenance of self-tolerance and prevention of autoimmunity. Blockade of the interaction between PD-L1 on tumor cells and PD-1 on T cells is expected to reverse T cell suppression within tumors. These agents are dependent on underlying T cell activation against the tumor cell to be effective.
- Avelumab is a fully human IgG1 anti-PDL1 antibody that selectively binds to PD-L1 and competitively blocks its interaction with PD-1.
- In ongoing phase 1 trials of avelumab, the agent has been well tolerated and has shown clinical activity.
- Clinical trials with anti-PD-1/L1 agents in colorectal cancer have resulted in minimal activity in patients who do not have mismatch repair deficiency (MMR-D).
- Therapeutic cancer vaccines targeting overexpressed proteins offer a potential method to activate T cells against tumors.
- A novel adenovirus based, CEA-targeting vaccine has demonstrated cytolytic T cell responses in patients with metastatic colorectal cancer.
- Standard of care agents in first line metastatic CRC have properties been associated with improved immune response via immunologic cell death and immunogenic modulation.

Primary Objective:

- To determine if there is an improvement progression free survival among patients with metastatic or unresectable colorectal cancer who are treated with standard of care + anti-PDL1 monoclonal antibody + Ad-CEA therapeutic cancer vaccine compared with standard of care alone.

Eligibility:

- Subjects age 18 and older with previously untreated pathologically confirmed metastatic or unresectable colorectal cancer; prior adjuvant therapy is acceptable.
- ECOG performance status ≤ 2.
- Normal organ and bone marrow function.
- Subjects with active autoimmune diseases requiring treatment and subjects requiring system steroids (except for physiologic doses for steroid replacement) are not allowed.
- Tumor sample and whole blood sample must be available for proteomics, genomics and transcriptomics analyses.
- Subjects with metastatic or unresectable colorectal cancer with mismatch repair deficiency (MMR-D or MSI-High) will not be eligible.

Design:

- This is a randomized, multicenter phase II clinical trial designed to evaluate the potential improvement in progression free survival (PFS) when Avelumab and Ad-CEA vaccine are used in combination with standard of care therapy in metastatic or unresectable colorectal cancer when compared with standard of care alone (FOLFOX-A).
- A lead in cohort, comprising the first 6 evaluable subjects enrolled, will be treated with avelumab + Ad-CEA vaccine + standard of care in order to assess the safety of the combination.
- If no more than 1 subject in the lead in cohort experiences a dose limiting toxicity attributable to the IND agents, 70 evaluable subjects will be randomized on a 1:1 basis to receive either Avelumab + Ad-CEA vaccine + standard of care (Arm B) or standard of care alone (Arm A)
- Standard of care therapy consists of 6 – 12 two week cycles of bevacizumab + FOLFOX (5-FU, leucovorin, oxaliplatin) followed by two week cycles of bevacizumab + capecitabine/5-FU until disease progression
- Subjects assigned to Arm A that have progressive disease will be offered Avelumab + Ad-CEA vaccine in combination with a standard chemotherapy regimen
- Kaplan-Meier curves and a two-tailed log-rank test will be the primary analysis methods.
- The accrual ceiling for the study is set at 97

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1 INTRODUCTION

1.1 STUDY OBJECTIVES

1.1.1 Primary Objective:

- To determine if there is an improvement in progression free survival among patients with metastatic or unresectable colorectal cancer who are treated with standard of care + Ad-CEA vaccine + Avelumab compared with standard of care alone

1.1.2 Secondary Objective:

- To determine the ratio of patients that are hospitalized because of adverse events attributed to disease progression

1.1.3 Exploratory Objective(s):

- Safety of the combination versus standard of care alone
- **Immunologic** effects of the combination compared with standard of care
    - **Peripheral**
        - Quantitate T cell responses against CEA, and other tumor associated antigens using methods such as intracellular cytokine staining, ELIspot, and/or others
        - Flow-based assay for analysis of PBMC subsets
        - T-cell clonal expansion assay
        - Serum cytokine and soluble factor analysis
        - Neutrophil lymphocyte ratio at baseline and change during treatment
        - HLA subtype correlation with clinical outcomes
        - ELISA for antibody generation against CEA
    - **Tumoral**
        - Immunohistochemistry analysis of T cell infiltrate (may include CD3, CD4, CD8, FoxP3 and others), immune regulatory markers (may include IDO, LAG3, TIM3, PD-1, PD-L1 and others)
        - RNA and proteomic analysis for:
            - immune signature changes
            - KRAS mutation status relationship to IL10 and TGF-beta tumor expression
            - Tumor subtyping into accepted molecular patterns
            - TLR4 loss of function polymorphisms and relationship to outcomes
        - Determine the genomic and proteomic profile of subjects' tumors to identify gene mutations, gene amplifications, RNA-expression levels, and protein-

expression levels. Correlations between genome/proteomic profiles and efficacy outcomes will be assessed.

- Correlative analysis of immune endpoints with clinical outcomes
- Overall response rate (CR+PR by RECIST 1.1)
- Overall survival

1.2 BACKGROUND AND RATIONALE

1.2.1 Metastatic colorectal cancer

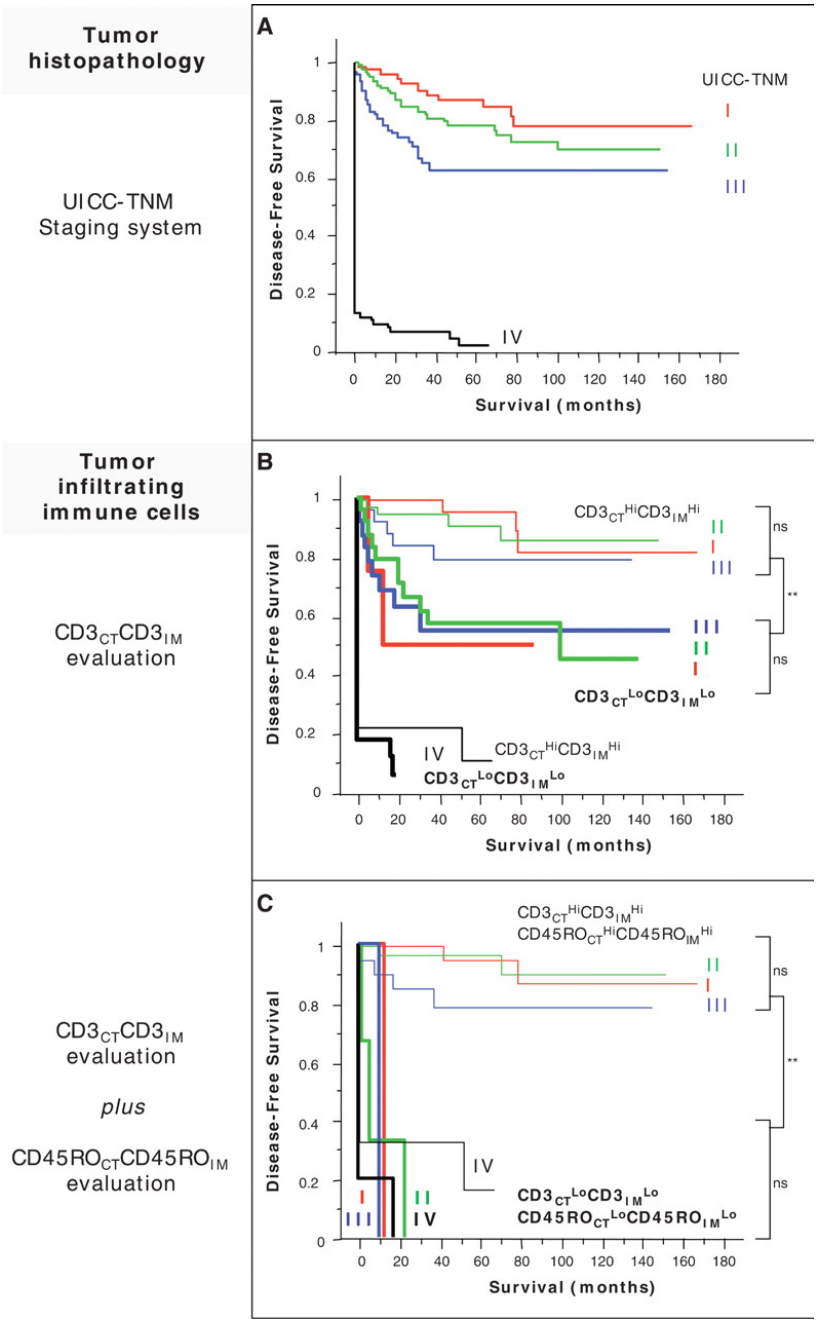
Colorectal cancer (CRC) is the third most common cancer diagnosis in the United States with a projected 132,700 new cases diagnosed in 2015. However, this disease accounts for the second most cancer-related deaths, projected to be 49,700 in 2015.[1]

1.2.2 Early stage colorectal cancer is immunologically modulated

Extensive work by the Galon, Pages, and Fridman group has identified, in retrospective analyses, immune cell tumor infiltrate signatures to be predictive of prognosis in early stage colorectal cancer at the time of diagnosis. Pages et al.[2] showed that signs of an immune response within colorectal cancers were associated with the absence of pathological evidence of early metastatic invasion and that an increased density of CD45RO+ memory T-cells was an independent predictor of increased overall survival (n = 490, P, 0.05). Five-year overall survival and disease-free survival were 46.3% and 43.1%, respectively, for tumors with a high density of CD45RO+ cells compared with 23.7% and 21.5% for tumors with a low density. Tumors without signs of early metastatic invasion had increased infiltrates of immune cells and increased levels of mRNA for products of TH1 effector cells, but not increased levels of inflammatory mediators or immunosuppressive molecules. Markers of T-cell migration, activation and differentiation were also increased, as well as the numbers of CD8+ T-cells. In addition, Galon et al.[3] further characterized the tumor infiltrating immune cells in the same cohort of colorectal cancer patients, and found that the type, density and location of the immune cells were a predictor of survival superior to the histopathological methods currently used, and independent of the tumor node metastasis (TNM) system (**Figure 1**). By conducting genomic and in situ immunostaining on resected tumors from patients with colorectal cancer, they found that TH1 adaptive immunity had a beneficial effect on clinical outcome. Tumors from patients without recurrence had significantly higher immune cell densities within both the center of the tumor (CT) and the invasive margin (IM). There was a statistically significant correlation between the immune cell density and patient outcome (n = 490, P, 0.05). They looked at all T-lymphocytes (CD3+), CD8+ T-cell effectors and memory T-cells (CD45RO+). Further investigation of the primary tumor microenvironment showed a correlation between the absence of metastasis to lymph nodes or distant organs and markers of innate immune cells (macrophages, dendritic cells, natural killer [NK] cells and NK T-cells) and activated T-cells.[4] The co-expression of genes for cytotoxicity and TH1 predicted patient survival independently of metastatic status (n = 142, P, 0.05). Pages et al.[5] recently published a study in which they classified early stage colorectal cancer patients (TNM I–II) into four different prognostic groups based on the density of CD45RO+ and CD8+ cells in different tumor regions. Six hundred and two tumors from two independent cohorts were investigated, and they found dramatic differences in disease-free, disease-specific and overall survival (**Figure 2**). Five-year survival in patients with high densities of both CD8+ and CD45RO+ cells was 86.2%, and only

4.8% of the patients had tumor recurrence, whereas in the group with low densities of these cells, 75% had tumor recurrence, and only 27.5% survived. The immune criteria were found to be independent prognostic factors in multivariate analysis (n = 602, P, 0.0001).

Figure 1. Disease free survival according to tumor histopathology (TNM staging), versus tumor infiltrating immune cell density ($CD3^+$ and $CD45RO^+$) in two separate tumor regions.

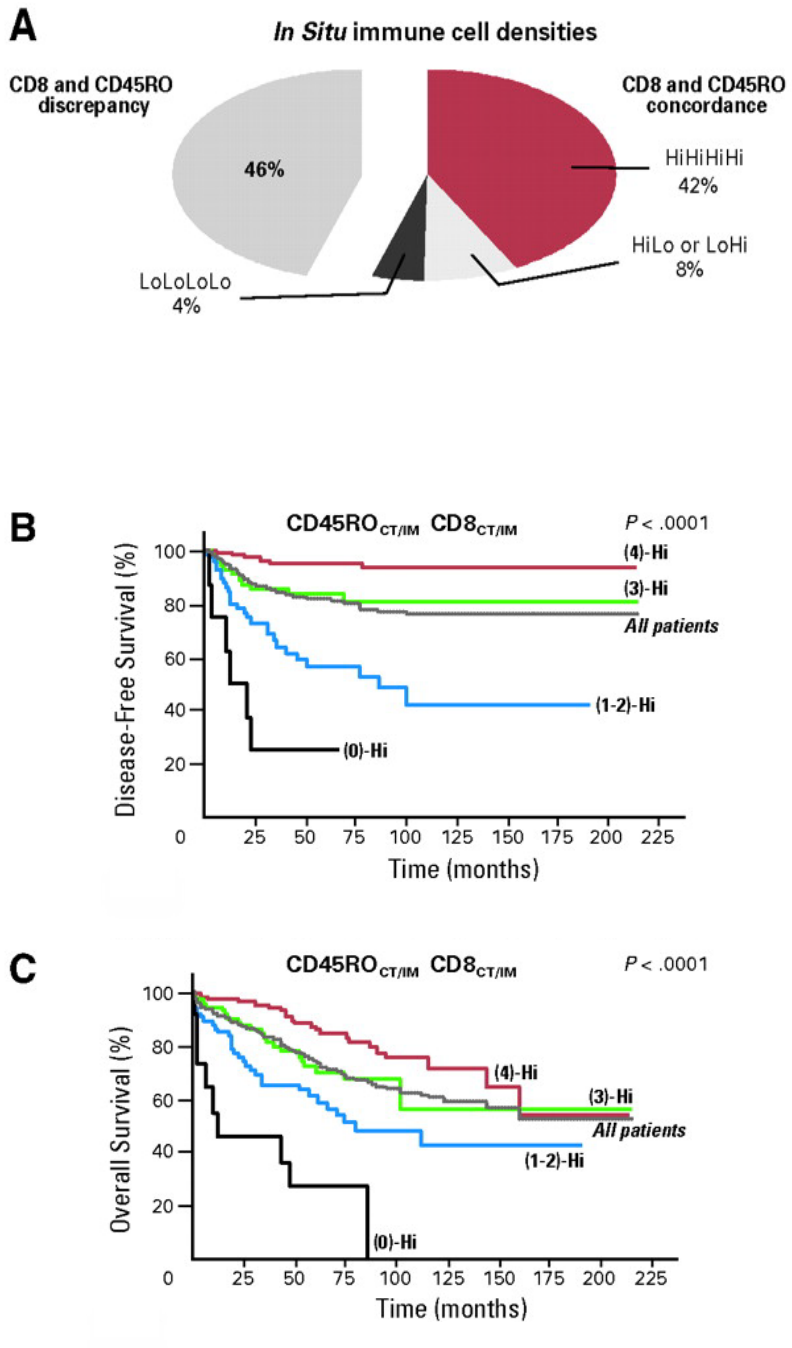


(A) Kaplan-Meier curves illustrate the duration of disease-free survival according to the UICC-TNM stages (stage I, red line [n = 75 patients]; stage II, green line [n = 137]; stage III, blue line [n = 99] and stage IV, black line [n = 95]) in patients with colorectal cancers.

(B) Kaplan-Meier curves illustrate the duration of disease-free survival according to the UICC-TNM stages (as in [a]) and according to the density of CD3+ cells in combined tumor regions ($CD3_{CT}{}^{Lo}CD3_{IM}{}^{Lo}$, thick lines, n=93 patients; $CD3_{CT}{}^{Hi}CD3_{IM}{}^{Hi}$, thin lines, n = 109). The subgroup of patients who did not appear to have a coordinated *in situ* immune reaction in tumor regions (Hi/Lo or Lo/Hi for CD3+ cell densities) presented Kaplan-Meier curves similar to those in the entire cohort.

(C) Kaplan-Meier curves illustrate the duration of disease-free survival according to the UICC-TNM stages and to the density of CD3+ and CD45RO+ cells in combined tumor regions ($CD3_{CT}{}^{Lo}CD3_{IM}{}^{Lo}$ plus $CD45RO_{CT}{}^{Lo}CD45RO_{IM}{}^{Lo}$, thick lines n = 16 patients; $CD3_{CT}{}^{Hi}CD3_{IM}{}^{Hi}$ plus $CD45RO_{CT}{}^{Hi}CD45RO_{IM}{}^{Hi}$, thin lines, n = 88). Cutoff values were 250, 640, 60 and 190 for $CD3_{CT}$, $CD3_{IM}$, $CD45RO_{CT}$ and $CD45_{IM}$, respectively. In (b and c), log-rank statistical test, **p < 10 4; ns, not significant; CT, center of the tumor; IM invasive margin.

Figure 2. Overall survival in Stage I and II CRC patients according to CD8+ and CD45RO+ cell densities.



(A) Pie chart illustrates the repartition of patients according to the presence of concordance (right part) or discrepancy (left part) between CD8+ and CD45RO+ cell densities in combined tumor regions of stages I and II colorectal tumors. Fifty-four percent of the patients presented with similar high (4)-Hi, low (0)-Hi or heterogeneous (2)-Hi immune infiltration for markers, whereas 46% presented with distinct patterns.

(B, C) Kaplan-Meier curves for the duration of disease- free survival and overall survival according to a combined analysis of CD8+ and CD45RO+ densities in tumor regions (center of the tumor [CT] and invasive margin [IM] in patients with stage I or II CRC. Patients are stratified according to an immune score ranging from 0 to 4, depending on the total number of high densities observed (two markers assessed in CT, two markers assessed in IM). For example, (4)-Hi refers to a tumor with high densities of CD8+ and CD45RO+ cells in CT and IM regions of the tumor (red line). Duration of disease-free survival and overall survival of the entire cohort of patients is also represented (gray line).

1.2.3 Advanced colorectal cancer is poorly immunogenic

While early stage disease appears to be immunologically relevant, the underlying immune response in advanced colorectal cancer appears to be very poor in the majority of patients with advanced colorectal cancer. This is indicated by a low number of PD-L1+ tumor cells despite the presence of CD8+PD-1+ T cells found in the intratumoral space in metastatic colorectal cancer cases.[6,7] This tumor microenvironment phenotype has been associated with a low likelihood of

anti-tumor effect when treated with PD-1/L1 blocking agents.[7] It has been hypothesized that a tumor microenvironment without PD-L1 expression indicates a poor T cell response against the tumor and that specific T-cell activation against target antigens in the tumor could lead to increased T cell infiltration with PD-1 expression and result in PD-L1 upregulation, which would make PD-1/L1 inhibition a viable anti-tumor strategy.[8] Previous work from our group has indicated that vaccination against target antigens overexpressed in the tumor can lead to significant infiltration of T cells in patients with prostate cancer.[9] Addition of therapeutic cancer vaccine capable of driving a specific T cell response to the tumor may overcome the poor immunogenicity of advanced colorectal cancer.

1.2.4 Ad5 [E1-, E2b-]-CEA(6D) vaccine (Ad-CEA vaccine)

1.2.4.1 Description of Ad-CEA vaccine

Overcoming the problem of pre-existing anti-vector immunity has been a subject of intense investigation. Investigations using other Ad subtypes or even non-human forms of Ad have been examined. Even if these approaches succeed in an initial immunization, subsequent vaccinations may be problematic due to immune responses generated against the differing Ad subtype. To overcome the Ad immunization barrier, and circumvent the adverse conditions for Ad5 [E1-] vectors, Etubics has constructed an advanced generation Ad vector. The new Ad5 [E1-, E2b-]-based vectors have additional deletions in the E2b region, removing the DNA polymerase and the preterminal protein genes. These vectors essentially do not replicate in cells, normal or transformed. These replication-defective vectors can only be propagated in the newly engineered, proprietary human 293 cell line (E.C7) that supplies the E1 and E2b gene functions *in trans* required for vector production.[10-12]

Deletion of the E2b region also confers advantageous immune properties on the novel Ad vectors. The deletion of E2b region sequences has the beneficial effect of markedly reducing residual late viral gene expression (>10,000 fold reduction in Ad-fiber protein levels).[10] This minimizes induction of anti-viral immune responses and allows for greater persistence of transduced cells. This enables more potent immune responses to specific, non-viral antigens while minimizing the immune responses to Ad viral proteins. Importantly, Ad5 [E1-, E2b-]-based vector vaccines can be injected multiple times in homologous immunization regimens designed to induce and increase specific immune responses to tumor antigens.[13-21] Ad-CEA is an Ad5 [E1-, E2b-]-based vector vaccine that contains the insertion of a modified CEA gene (CEA(6D)) designed to increase cell mediated immune (CMI) responses to CEA in immunized patients.[22-24]

1.2.4.2 Preclinical data

In pre-clinical studies,[16] the immunogenicity of Ad5 [E1-, E2b-]-CEA(6D) vaccine was tested in an Ad5 naïve and an Ad5 immune murine model. To assess the induction of cellular meditated immunity following immunization, Ad5 naïve C57Bl/6 mice were immunized three times subcutaneously (SQ) with 10^{10} VP of Ad5 [E1-]-CEA(6D) at weekly intervals. Three immunizations were used because our prior studies in an HIV-1 model demonstrated that the Ad5 [E1-, E2b-] vector induced a maximum immune boost after three immunizations.[17,18] Fourteen days after the final immunization, CEA-specific CMI responses were determined by IFN-γ and IL-2 ELISpot assays. Mice immunized with Ad5 [E1-, E2b-]-CEA(6D) induced a significantly greater number of IFN-γ ($p<0.01$) and IL-2 ($p<0.01$) secreting splenocytes than mice immunized with Ad5 [E1-]-CEA(6D) (**Figure 3 A, B**). Splenocytes from vaccinated mice and controls were assessed

for non-specific cytokine secretion following stimulation with HIV-1 Gag and β-galactosidase. Non-specific IFN-γ or IL-2 secretion was not detected in T-cells from the spleens of mice vaccinated with either CEA expressing vector (**Figure 3**). Splenocytes were also stimulated with Ad5-null to confirm positive vaccination against Ad5 and splenocytes derived from all such immunized mice harbored Ad5 specific T-cells (**Figure 3**). Elevated levels of CEA specific IgG antibody were detected after immunizations and these levels were comparable for both Ad5-CEA vectors.

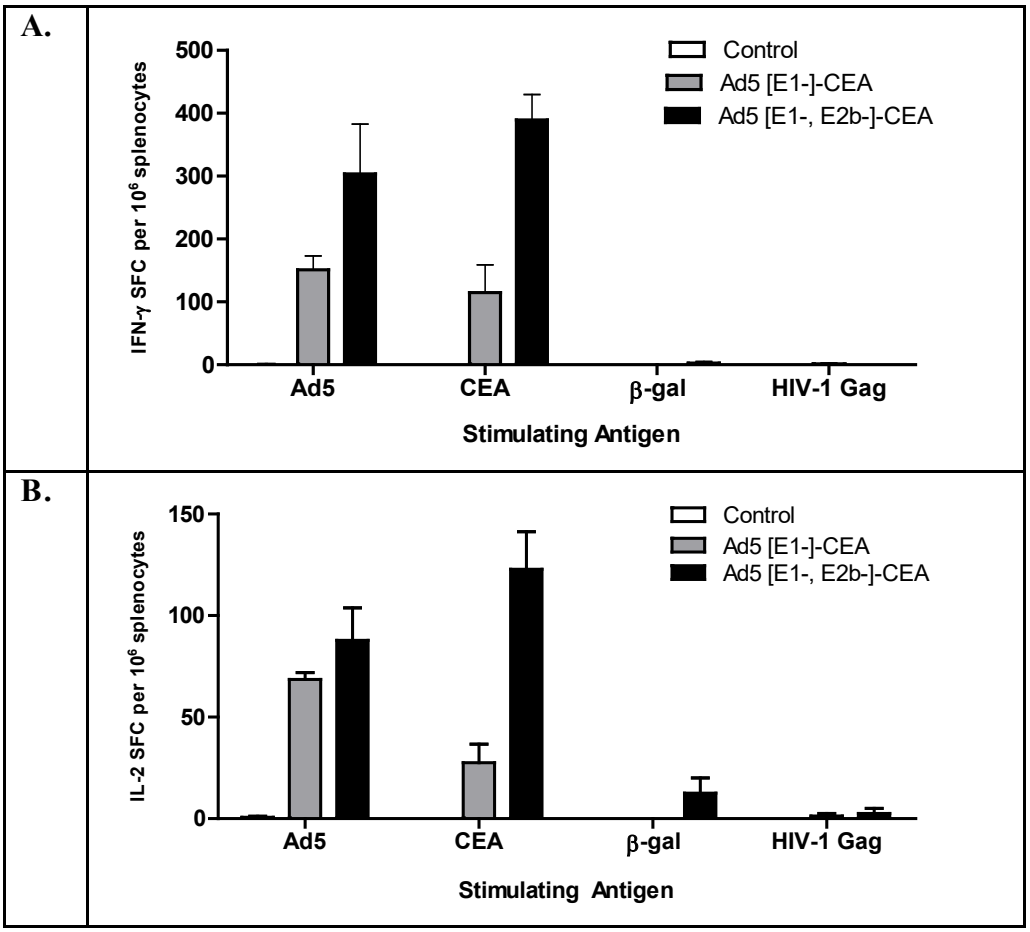


Figure 3. Immunization with Ad5-CEA(6D) platforms.

Ad5 naïve C57Bl/6 mice (n=7/group) were immunized three times at one week intervals with 10^{10} VP of Ad5 [E1-]-CEA(6D), Ad5 [E1-, E2b-]-CEA(6D), or injection buffer alone (control). Fourteen days after the final immunization splenocytes were assessed for the number of IFN-γ secreting T-cells (A) or the number of IL-2 secreting T-cells (B) by ELISpot analysis. Splenocytes were also assessed for non-specific IFN-γ or IL-2 secreting T cells by stimulation with the non-immunizing antigens β-galactosidase (β-gal) and HIV-1 Gag. There was a significantly ($P<0.05$) greater level of IFN-γ and IL-2 secreting T-cells in mice that were immunized with Ad5 [E1-, E2b-]-CEA(6D). The error bars depict the SEM.

To test the hypothesis that Ad5 [E1-, E2b-]-CE(6D) is more effective than Ad5 [E1-]-CEA(6D) in the presence of Ad5 immunity, Ad5 naive C57Bl/6 mice were immunized twice with 10^{10} VP of Ad5 [E1-]-null to induce Ad5 immunity. The presence of anti-Ad5 IgG antibody was confirmed

by ELISA as 0.028 ± 0.028 ng Equivalents of IgG antibody for baseline samples versus 39.50 ± 3.78 ng Equivalents of IgG antibody in samples after Ad5-null immunizations ($p<0.0001$). Ad5 immune mice were then immunized three times at weekly intervals with 10^{10} VP of Ad5 [E1-]-CEA(6D) or Ad5 [E1-, E2b-]-CEA(6D). Splenocytes were collected 14 days after the final immunization and assessed by IFN-γ and IL-2 ELISpot. Ad5-immune mice immunized with Ad5 [E1-, E2b-]-CEA(6D) exhibited significantly higher levels of IFN-γ ($p=0.04$) and IL-2 ($p<0.01$) secreting splenocytes as compared to Ad5 immune mice immunized with Ad5 [E1-]-CEA(6D) (**Figure 4**). When compared to the results for naïve mice above, levels of CEA specific IgG antibody generated after immunizations were greatly reduced.

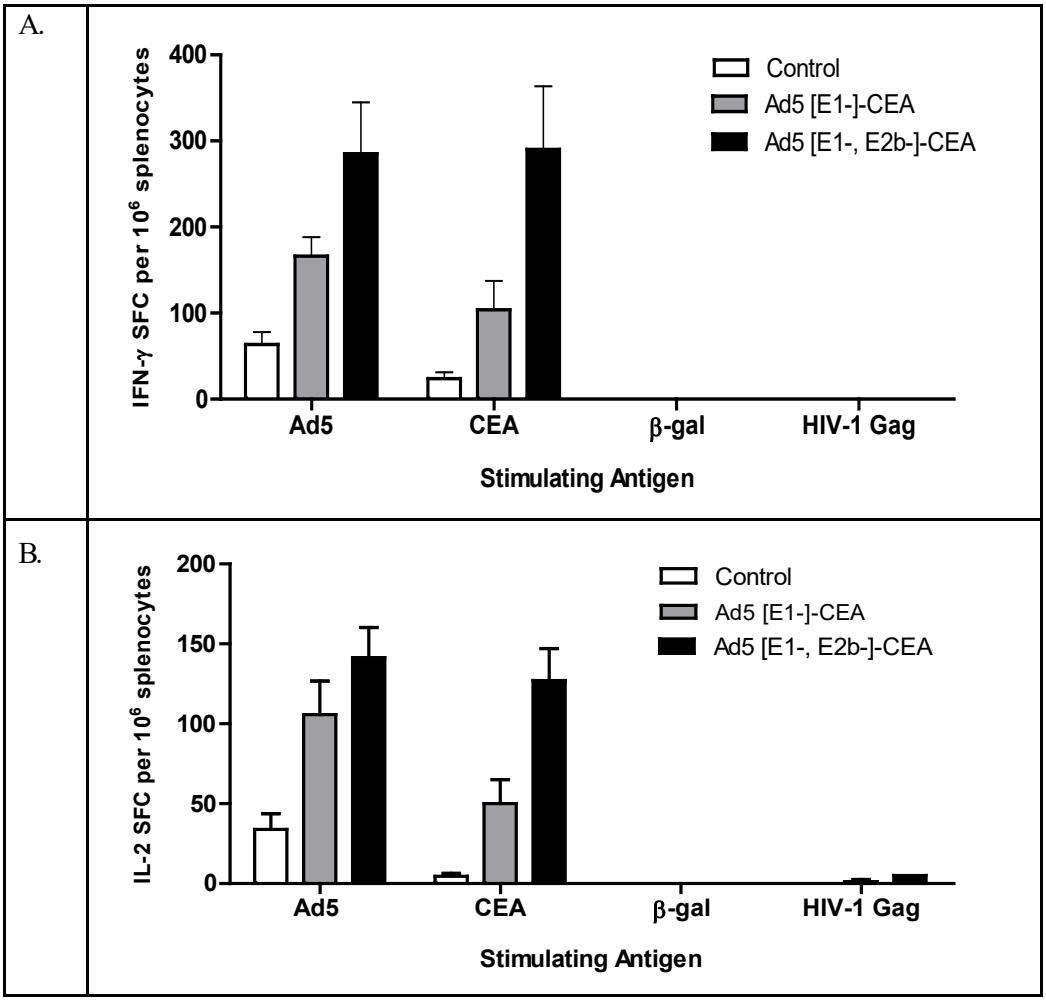


Figure 4. CMI against Ad5 [E1-]-CEA(6D) and Ad5 [E1-, E2b]-CEA(6D) in Ad5 immune mice.

C57Bl/6 mice (n=7/group) were pre-immunized two times with 10^{10} VP Ad5-null to induce Ad5 immunity. They were then immunized three times at one week intervals with 10^{10} VP Ad5 [E1-]-CEA(6D) or Ad5 [E1-, E2b-]-CEA(6D). Controls received injection buffer alone. Splenocytes were collected 14 days after the final immunization and assessed for IFN-γ secreting T-cells (A) or IL-2 (B) by ELISpot assay. Splenocytes were also assessed for non-specific secreting T-cells of IFN-γ (A) or IL-2 (B) by stimulation with the non-immunizing antigens Beta-galactosidase (β-gal) and HIV-1 Gag. Note the significantly ($p<0.05$) greater levels of IFN-γ and IL-2 secreting T-cells in mice that were immunized with Ad5 [E1-, E2b-]-CEA(6D). The error bars depict the SEM.

Having immunologically compared the two Ad5-CEA(6D) vectors, a study was then performed to determine if the Ad5 [E1-, E2b-]-CEA(6D) vector could break tolerance in the more stringent CEA transgenic mouse model in which CEA is a self-antigen. Mice were immunized once with 2.6 X 10^{10} VP of Ad5 [E1-, E2b-]-CEA(6D) and 14 days later, their splenocytes were assessed for CMI responses by IFN-γ ELISpot assays. As presented in **Figure 5**, elevated numbers of IFN-γ secreting splenocytes were observed after only one injection with the vector platform. ELISpot studies employing Cytomegalovirus (CMVpp65) and HIV-gag antigens demonstrated that the response was specific to CEA.

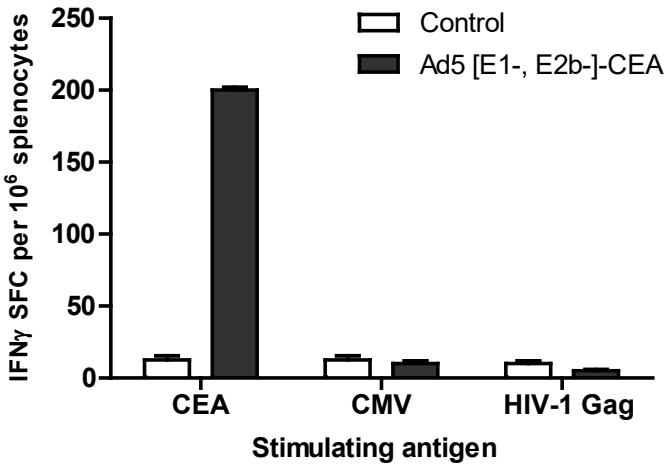


Figure 5. Ability of the Ad5 [E1-, E2b-]-CEA(6D) vector to break tolerance.

C57Bl/6 mice transgenic for CEA were immunized once with Ad5 [E1-, E2-]-CEA(6D) and tested for CMI response activity to CEA. Note the elevated levels of IFN-γ secreting splenocytes specific for CEA and not for irrelevant antigens such as CMV or HIV-1 Gag. The error bars depict the SEM.

The efficacy of immunizations with Ad5 [E1-, E2b-]-CEA(6D) was evaluated by the treatment of CEA expressing MC-38 tumors that were growing in Ad5 naïve and Ad5 immune mice. A group of C57Bl/6 mice (n=7) were implanted SQ with 10^6 CEA expressing MC-38 tumor cells (MC38-CEA). Six days later, when tumors were palpable (3-5mm), the mice were treated with three SQ injections of 10^{10} VP of Ad5 [E1-, E2b-]-CEA(6D) on days 7, 13 and 19. Untreated C57Bl/6 mice (n=7) implanted with 10^6 MC38-CEA cells and injected at the same time points with injection buffer alone served as controls. Tumor volumes were measured and reported as previously described.[16] The sizes of treated and control tumors are presented in **Figure 6A** as a function of time. At Day 20 post implant the tumors in mice treated with Ad5 [E1-, E2b-]-CEA(6D) were significantly ($p<0.05$) smaller as compared to controls (**Figure 6A**). Tumors were excised and weighed at the termination of the study. Tumors from mice treated with Ad5 [E1-, E2b-]-CEA(6D) weighed significantly ($p<0.05$) less than tumors from control mice (**Figure 6B**). Splenocytes from both groups of mice were assessed for the number of CEA stimulated IFN-γ secreting T cells by ELISpot. Mice immunized with Ad5 [E1-, E2b-]-CEA(6D) had a significantly ($p<0.05$) greater number of INF-γ secreting T-cells in their spleens as compared to control mice (**Figure 6C**).

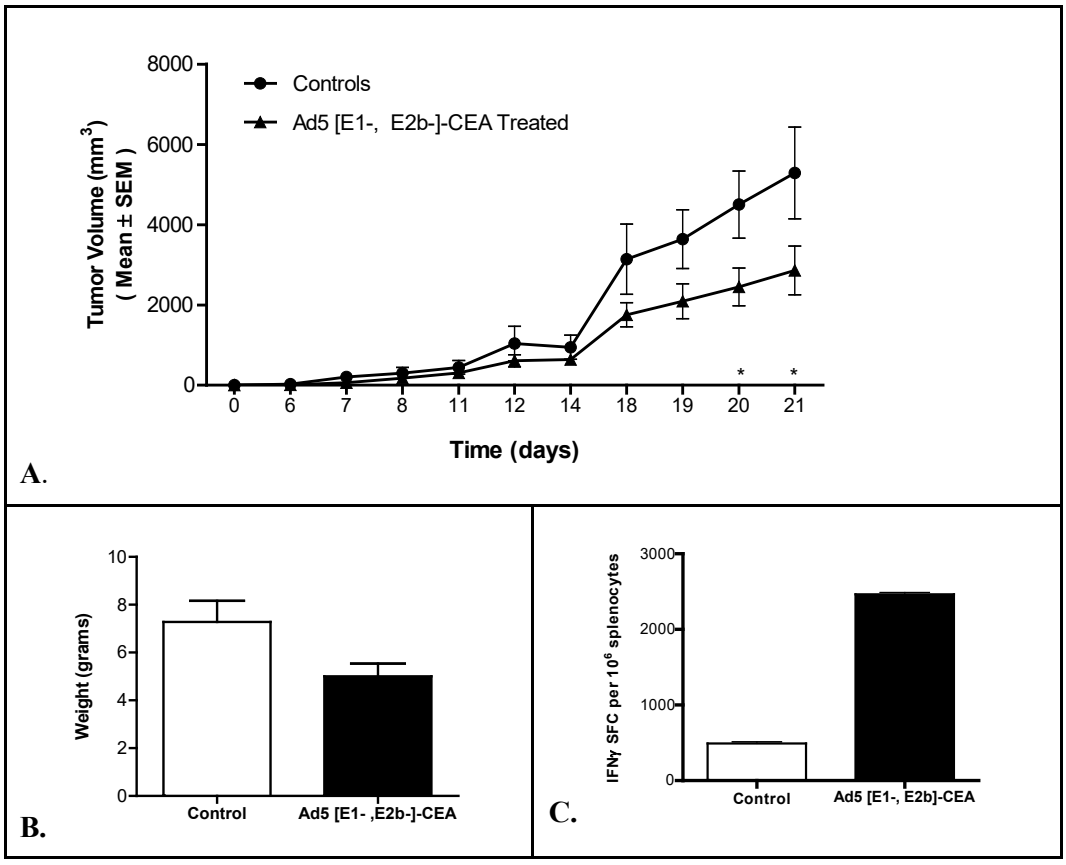


Figure 6. Treatment of CEA expressing tumors in Ad5 naïve mice.

C57Bl/6 mice (n=7/group) were implanted in the flank with 10^6 MC-38 CEA expressing tumor cells and subsequently treated with three immunizations of 10^{10} VP of Ad5 [E1-, E2b-]-CEA vaccine at one week intervals. Tumors were measured by two opposing diameters and reported as a multiple of those numbers to determine tumor volumes (A). Statistical differences were determined using the Bonferroni posttests analysis with PRISM software. * Indicates a significant difference in the value of the means on that day. Tumors were excised at the termination of the study and weighed. Tumors from mice treated with Ad5 [E1-, E2b-]-CEA(6D) weighed significantly less ($p<0.05$) (B). Splenocytes were collected and assessed for T-cells which secreted IFN-γ by ELISpot analysis and the data is presented. Note the significantly ($p<0.05$) greater levels of IFN-γ and IL-2 secreting T-cells in mice that were immunized with Ad5 [E1-, E2b-]-CEA(6D) (C). Note that there was a significant reduction in tumor size in treated mice at days 20 & 21 as compared to untreated controls. Values represent Mean ± SEM.

To determine if Ad5 immune MC38-CEA tumor bearing mice could be treated with Ad5 [E1-, E2b-]-CEA(6D), C57Bl/6 mice (n=7) were immunized two times at a two-week interval with 10^{10} VP of Ad5 [E1-]-null. Two weeks after the second Ad5 [E1-]-null immunization, mice were implanted SQ with 10^6 CEA expressing MC-38 tumor cells. Six days later tumors were measurable (3-5mm) and the mice were treated with three SQ injections of 10^{10} VP of Ad5 [E1-, E2b-]-CEA(6D) at weekly intervals. C57Bl/6 mice (n=7) which were immunized twice with Ad5 [E1-]-null, inoculated with 10^6 MC38-CEA cells and injected three times SQ with injection buffer alone served as controls. All mice were monitored for tumor growth and tumor volumes are reported using previously described methods.[16] Evaluation was over a 21-day period and the size of MC38-CEA tumors was determined as a function of time. At day 19 the tumors in mice treated with Ad5 [E1-, E2b-]-CEA(6D) were significantly smaller as compared to the controls (**Figure 7A**). At the

end of the study all mice were sacrificed and the tumors were excised and weighed. Tumors from the mice treated with Ad5 [E1-, E2b-]-CEA(6D) weighed significantly less than the tumors from controls (**Figure 7B**). The number of CEA stimulated T-cells in the spleen which secreted IFN-γ was determined by ELISpot assay. As presented in **Figure 7C**, mice immunized with Ad5 [E1-, E2b-]-CEA(6D) had a significantly greater number of IFN-γ secreting T-cells than did control mice that received injection buffer alone.

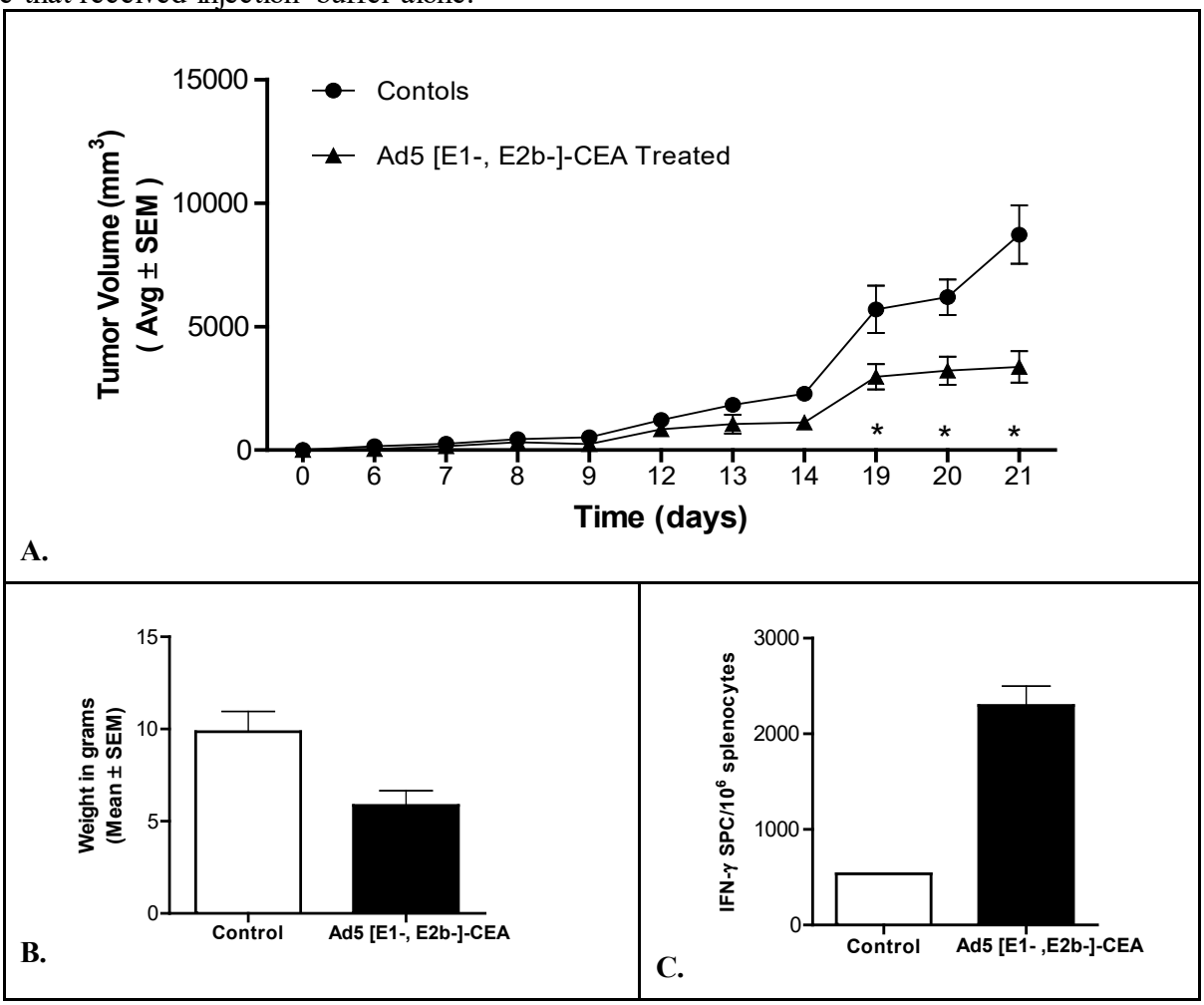


Figure 7. Treatment of CEA expressing tumors in Ad5 immune mice.

C57Bl/6 mice (n=7/group) were immunized twice at fourteen-day intervals with 10^{10} VP of Ad5 [E1-]-null. Fourteen days later, mice were implanted with 10^6 MC-38 CEA expressing tumor cells in the flank and subsequently treated with three immunizations of 10^{10} VP of Ad5 [E1-, E2b-]-CEA(6D) vaccine weekly. Tumors were measured by two opposing diameters and a multiple of the numbers determined the tumor volume. Statistical differences were determined using the Bonferroni posttests analysis with PRISM software. Note that there was a significant reduction in tumor size in treated mice at days 20 & 21 as compared to untreated controls (A). * Indicates a significant difference in the value of the means on that day. Tumors were excised at the termination of the study and weighted. Tumors from mice treated with Ad5 [E1-, E2b-]-CEA(6D) weighed significantly less ($p<0.05$) (B). Splenocytes were collected and assessed for the number of T-cells which secreted IFN-γ by ELISpot. Note the significantly ($p<0.05$) greater levels of IFN-γ and IL-2 secreting T-cells in mice that were immunized with Ad5 [E1-, E2b-]-CEA(6D) (C). Values represent Mean ± SEM.

In summary, Ad5 [E1-, E2b-]-CEA(6D) is a therapeutic vaccine targeting CEA that induces robust immune responses, even in the presence of pre-existing Ad5 immunity. When compared directly with a conventional Ad5 [E1-]-CEA(6D) vector, the Ad5 [E1-, E2b-]-CEA(6D) platform induced superior cellular mediated immune responses in both Ad5 naïve and Ad5 immune animals. In therapeutic efficacy studies, treatment with Ad5 [E1-, E2b-]-CEA(6D) resulted in significantly decreased established MC38-CEA tumor growth even in mice which had been pre-immunized against Ad5. Treatment with Ad5 [E1-, E2b-]-CEA(6D) did not result in any adverse effects in pre-clinical studies. Our goal is to further develop this therapeutic vaccine against CEA and utilize this novel Ad5 vector system that overcomes barriers found with other Ad5 systems. The results of these pre-clinical studies establish the technical and immunological merit of using the Ad5 [E1-, E2b-]-CEA(6D) vaccine as a therapeutic agent.

1.2.4.3 Clinical safety

Etubics Corporation has performed a Phase I/II clinical trial of Ad-CEA (Ad5 [E1-, E2b-]-CEA(6D)) (IND#14325).[13,19] The Phase I/II study consisted of a dose-escalation study of four dosage levels ($1x10^9$, $1x10^{10}$, $1x10^{11}$, $5x10^{11}$ VP/dose) of ETBX-011 (Phase I component), and the maximally tolerated dose of ETBX-011 (Phase II and $5x10^{11}$ VP/dose components). Ad-CEA was administered by SQ injection every 3 weeks. Thirty-two patients with metastatic colorectal cancer, median age 57.5 (range 38–77) who had failed a median of three prior chemotherapeutic regimens (range 2–5), had a performance status of 90 % (range 70–100 %), and had three sites of metastatic disease (range 1–4), were enrolled. The majority of patients were able to receive all three immunizations. Four patients who stopped immunizations early did so due to significant disease progression.

A total of 94 immunization treatments were administered to all patients. There was no dose-limiting toxicity and no serious adverse effects (SAE) that resulted in treatment discontinuation at any vaccine dose level. The most common toxicity was a self-limited, injection site reaction. Other reactions occurred with less than a 10% incidence of all adverse effects (AE) reported and included fever, flu-like symptoms, anorexia, chills, nausea, and headache. These symptoms were also self-limiting and did not require intervention other than symptomatic measures such as acetaminophen.[13,19]

1.2.4.4 Clinical immune response

A secondary objective was to evaluate CEA specific immune responses following immunization treatments with the product candidate. As determined by an ELISA technique,[13,19] we observed no antibody activity directed against CEA. We assessed CMI responses in colorectal cancer patients treated in cohort 1, cohort 2, cohort 3/Phase II, and cohort 5. PBMCs were isolated prior to immunotherapy treatment and after all treatments as well as three weeks following the last treatment from patients. CEA specific ELISpot assays were performed on PBMC as previously described[13,16,19] to determine the numbers of interferon gamma (IFN-γ) secreting lymphocytes (SFC) after exposure to CEA peptides *in vitro*. We determined the highest CMI responses during immunizations, regardless of time point (weeks 3, 6, or 9) in the patients treated in cohort 1, cohort 2, cohort 3/Phase II, and cohort 5. This analysis revealed a dose response to increasing levels of product (**Figure 8**). The highest CMI levels occurred in patients that received the highest dose of $5x10^{11}$ VP (Cohort 5). In a preliminary study, we observed a population of polyfunctional CD8+ T cells (those that secrete more than 1 cytokine when activated) after immunizations that secreted multiple cytokines, a sign of greater functionality of T cells induced by the vaccine. In further

follow-up analysis of a few patient blood samples, we noted a decrease in CEA directed immune responses after immunizations were stopped (**Figure 9**). This observation supports a rationale for booster immunizations.

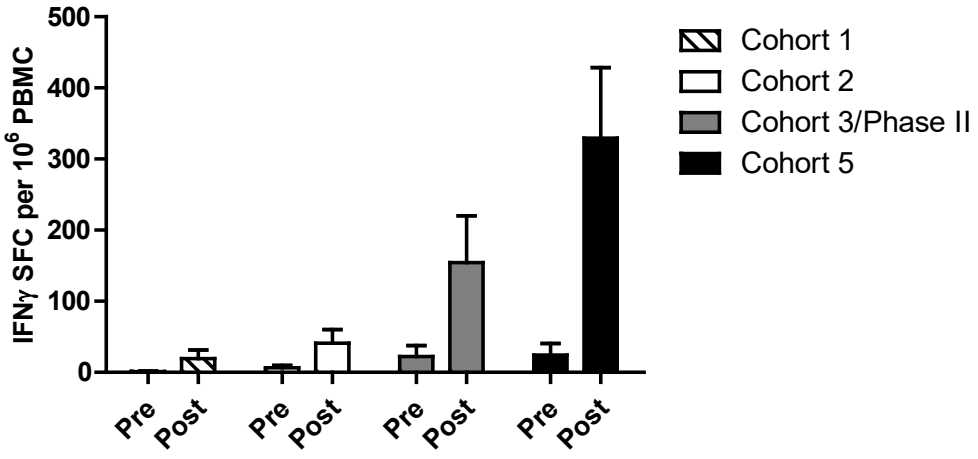


Figure 8. CMI responses in treated patients. CMI (IFN-γ secretion) was assessed at baseline (Pre) and after administrations of Ad5 [E1-, E2b-]-CEA(6D) (Post). The highest CMI responses (regardless of time point) observed in the patients after treatment revealed a dose response. The highest CMI levels occurred in patients that received the highest dose of $5x10^{11}$ VP (Cohort 5). The CMI responses for Cohort 3/Phase II and Cohort 5 were significantly elevated (P=0.0002 and P=0.0317, respectively; *Mann-Whitney test*) as compared to their baseline (Pre) values. Specificity of the responses was demonstrated by the lack of reactivity with the irrelevant antigens β-galactosidase and HIV-gag (data not shown). For positive controls, PBMCs were exposed to concanavalin A (data not shown). SFC: spot forming cells. Values = Mean ± SEM for each Cohort.

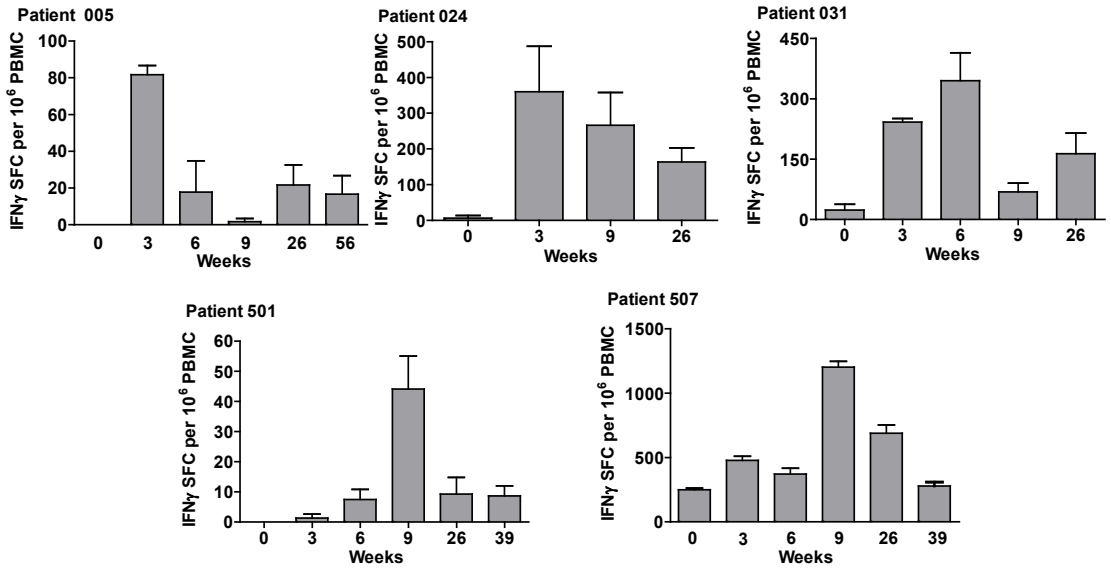


Figure 9. Profiles of CEA directed CMI responses in 5 mCRC patients during and after the course of immunotherapy. Note the decrease in CMI responses from their peak values after immunizations ended. Values are Mean ± SEM.

We also measured Ad5 NAb and CMI against Ad5 and correlated it with CEA-specific CMI. Each patient had their serum and PBMC sample tested at baseline (prior to treatment) and at 9 weeks after completion of 3 treatments. Nineteen of 31 colorectal cancer patients (61%) tested in this study had Ad5 neutralizing activity in serum samples prior to the onset of treatment with Ad5 [E1-, E2b-]-CEA(6D). The mean pre-treatment Ad5 NAb titer value obtained among all patients was 1:189 ±1:71 SEM and the mean pre-treatment Ad5 NAb titer among seropositive patients was 1:308 ± 1:108. Analysis of serum samples from patients who received 3 immunizations revealed Ad5 NAb titers that were significantly increased ($P<0.0001$, *Mann-Whitney test*) by week 9 (mean 1:4767 ± 1:1225 SEM) when compared with their respective baseline values. Analysis of PBMC for CMI responses to Ad5 also revealed a significant increase ($P<0.01$, Mann-Whitney test) in Ad5 directed CMI responses after immunizations with Ad5 [E1-, E2b-]-CEA(6D) (22.6 ± 9.3 SEM IFN-γ secreting SFC at week 0 versus 191.1 ± 83.7 IFN-γ SFC at week 9).

1.2.4.5 Clinical evidence of activity

The Ad-CEA treated colorectal cancer patients (total=32) were followed for survival and Kaplan-Meier plots and survival proportions performed (PRISM software).[13,19] Events were determined by information from the social security death index (SSDI) database, clinical charts and telephone calls (**Figure 10**).

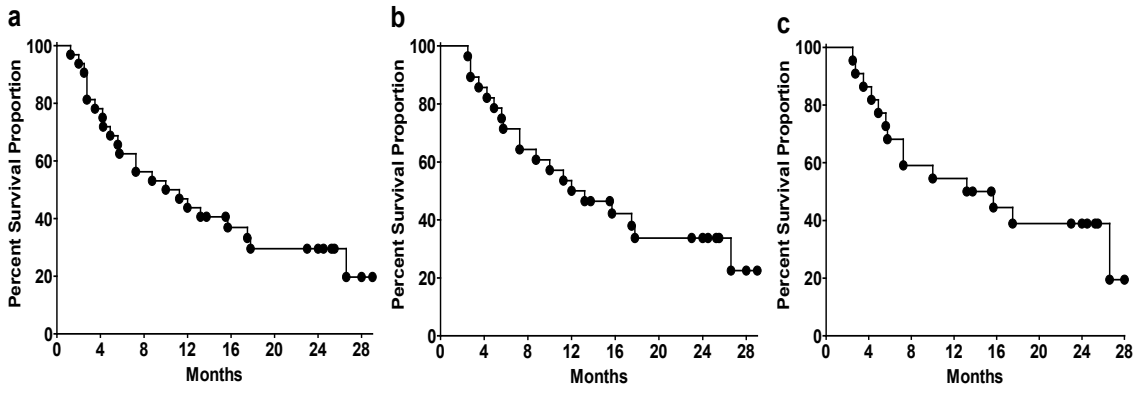


Figure 10. Kaplan-Meier survival plots on long-term overall survival of treated mCRC patients. Panel a represents all treated patients. Panel b represents patients that received all 3 treatments. Panel c represents patients immunized 3 times with the 2 highest doses of vaccine. There were 23 events **during the study.**

The seven patients in cohorts 1 and 2 experienced a 12-month survival proportion of 29%. The 21 patients in cohort 3 and Phase II experienced a 12-month survival proportion of 48%. The six patients in cohort 5 experienced a 12-month survival proportion of 50%. Twenty-nine month overall survival of the intent-to-treat population (32 patients) was 20% (**Figure 10A**) with a median survival time of 11 months from informed consent/first injection. For the subset of 28 patients that received all 3 immunizations, the 29-month survival was 23% (**Figure 10B**) with a median survival time of 13 months. For the 22 patients optimally dosed with the two highest doses of vaccine (1 and 5 x 10^{11}) and receiving all 3 immunizations, the 28-month overall survival was 19% (**Figure 10C**). Median overall survival was 13 months in the optimally treated patients. Since there was no active control group in the study, comparisons for significance in survival time cannot be made. There were 3 stable disease events observed immediately after completion of treatment.

1.2.4.6 Evidence of an Ad5 [E1-, E2b-]-based vaccine driving T cell infiltration and PD-L1 upregulation

High-risk human papillomavirus (HPV) such as HPV type-16 is associated with the etiology of cervical and more than 90% of HPV-related head and neck squamous cell carcinomas (HNSCC).[25-27] Preventive vaccines such as Human Papillomavirus Bivalent [Types 16 and 18] Vaccine and Human Papillomavirus Quadrivalent [Types 6, 11, 16, and 18] Vaccine are currently being used as a primary defense against HPV associated cancers by preventing infection with the virus but reports indicate that they are not effective for active immunotherapy of established disease.[28] Immunotherapeutic treatment of HPV induced malignances has been investigated by us and others. The HPV early 6 (E6) and early 7 (E7) genes are expressed at high levels in HPV-induced cancers and are involved in the immortalization of primary human epidermal cells.[26,28-32] Thus, these are ideal targets for tumor-specific immunotherapy because unlike many other tumor-associated antigens these viral antigens are "non-self" and thus do not have the potential to induce autoimmunity.[21,28-33]

Using the Ad5 [E1-, E2b-]-based vector platform, we investigated in a murine model of HPV-associated E6/E7 expressing cancer if immunotherapy against human papilloma virus (HPV) using a viral gene delivery platform to immunize against HPV 16 genes E6 and E7 (Ad5 [E1-, E2b-]-E6/E7) combined with programmed death-ligand 1 (PD-1) blockade could increase therapeutic

effect as compared to the vaccine alone.[33] Ad5 [E1-, E2b-]-E6/E7 as a single agent induced HPV-E6/E7 cell-mediated immunity. Immunotherapy using Ad5 [E1-, E2b-]-E6/E7 resulted in clearance of small tumors and an overall survival benefit in mice with larger established tumors. When immunotherapy was combined with immune checkpoint blockade, an increased level of anti-tumor activity against large tumors was observed (**Figure 11**). Analysis of the tumor microenvironment in Ad5 [E1-, E2b-]-E6/E7 treated mice revealed elevated $CD8^+$ tumor infiltrating lymphocytes (TILs); however, we observed induction of suppressive mechanisms such as programmed death-ligand 1 (PD-L1) expression on tumor cells and an increase in $PD\text{-}1^+$ TILs. When Ad5 [E1-, E2b-]-E6/E7 immunotherapy was combined with anti-PD-1 antibody, we observed $CD8^+$ TILs at the same level but a reduction in tumor PD-L1 expression on tumor cells and reduced $PD\text{-}1^+$ TILs providing a mechanism by which combination therapy favors a tumor clearance state and a rationale for pairing antigen-specific vaccines with checkpoint inhibitors in future clinical trials.

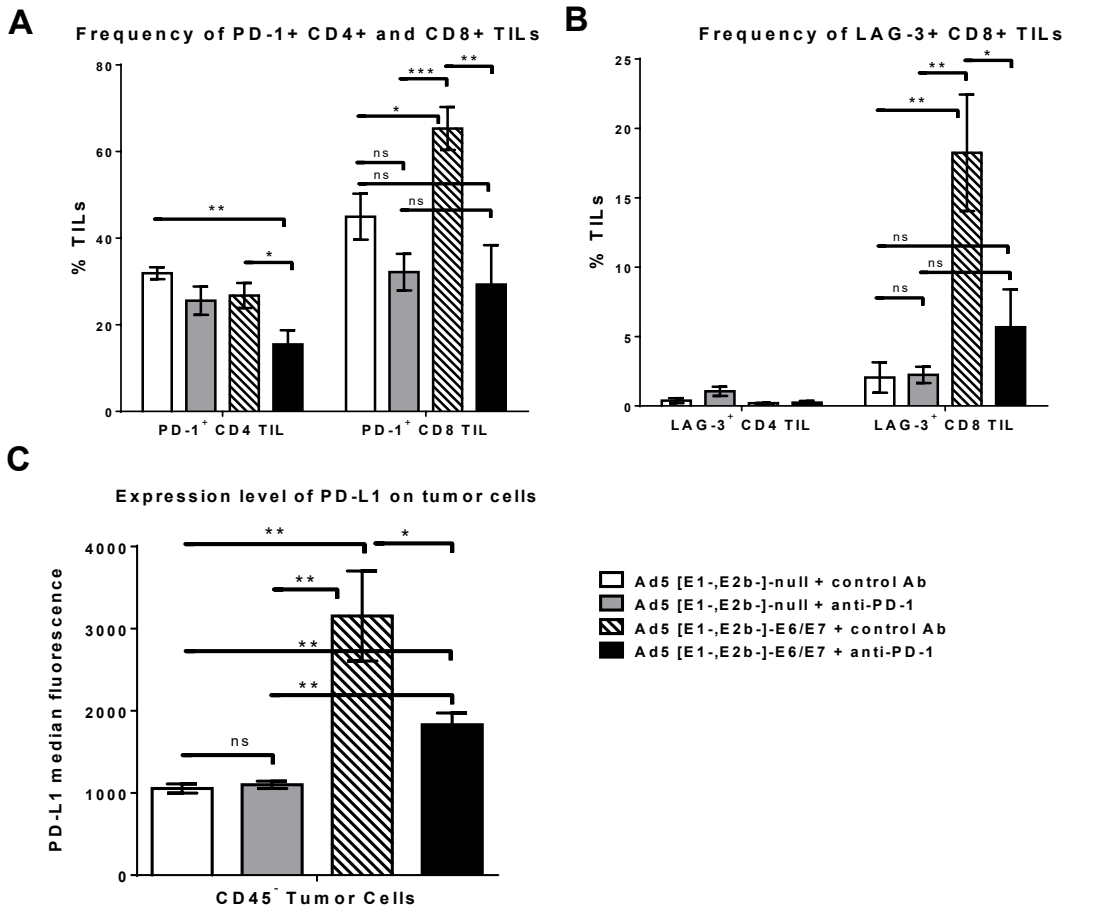


Figure 11. Ad5 [E1-, E2b-]-E6/E7 plus anti-PD-1 combination therapy promotes a pro-inflammatory tumor microenvironment. C57BL/6 mice (n=5/group) were tumor implanted, treated, and tumors were analyzed. The frequency of PD-1$^+$ CD4$^+$ and CD8$^+$ TILs (A), LAG-3$^+$ CD8$^+$ TILs (B), and expression level of PD-L1 on tumor cells (C) is increased in tumors from mice treated with Ad5 [E1-, E2b-]-E6/E7. Tumors from mice treated with a combination of Ad5 [E1-, E2b-]-E6/E7 and anti-PD-1 have a significantly lower frequency of PD-1$^+$ CD4$^+$ and CD8$^+$ TILs (A), a significantly lower frequency of LAG-3$^+$ CD8$^+$ TILs (B), and the expression level of PD-L1 on the tumor cells significantly reduced (C), **bringing these levels more in line with tumors from control mice. Significance analysis was**

1.2.4.7 Rationale for combining chemotherapy treatment with Ad5 [E1-, E2b-]-based vaccine immunotherapy

Using the same murine model of HPV-associated E6/E7 expressing cancer described above (see section **1.2.4.6**), we investigated if immunotherapy using Ad5 [E1-, E2b-]-E6/E7 could be combined with cisplatin/radiation treatment that is utilized to treat HNSCC.[21] We observed that when immunotherapy was combined with cisplatin/radiation treatment, a significant increase in survival time demonstrated in tumor bearing mice treated with the combination of immunotherapy

and cisplatin/radiation treatment as compared to control mice receiving cisplatin/radiation treatment alone (**Figure 12**).

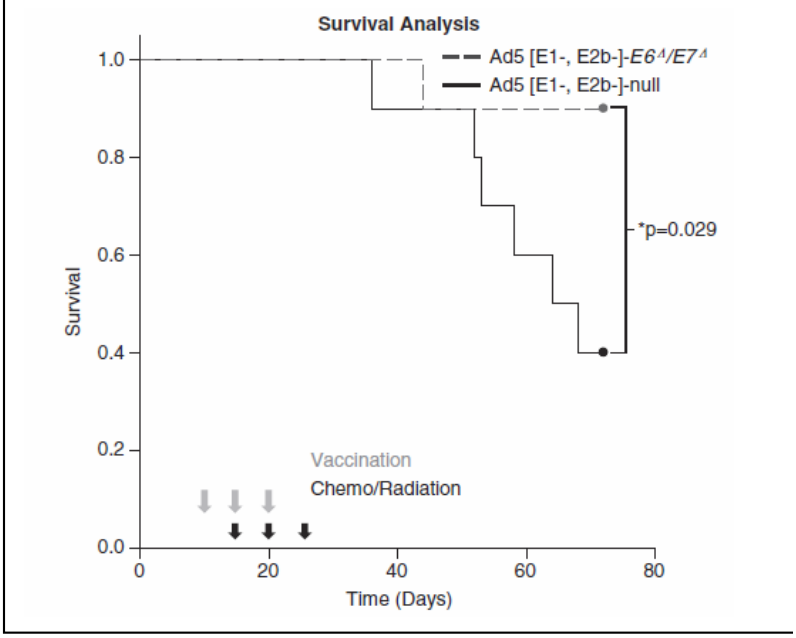


Figure 12. Immunotherapy combined with chemotherapy/ radiation treatment of established HPV-E6/E7 expressing tumors. Established HPV-E6/E7 expressing tumors were treated by immunotherapy on days 7, 14, & 21 combined with cisplatin/radiation treatment on days 13, 20, & 27. Control tumor bearing mice were treated by injections with Ad-null combined with cisplatin/radiation treatment.

In light of the above results, we then investigated the effects of combining immunizations with Ad5 [E1-, E2b-]-E6/E7 with cisplatin/radiation treatment versus cisplatin/radiation treatment alone in a murine model. We observed that the combination of Ad5 [E1-, E2b-]-E6/E7 immunizations plus cisplatin/radiation treatment resulted in the induction greater CMI responses as compared to immunizations with Ad5 [E1-, E2b-]-E6/E7 alone (**Figure 13**). These results provide us with a rationale for combining immunotherapy with chemotherapy treatment in order to achieve greater anti-tumor CMI responses.

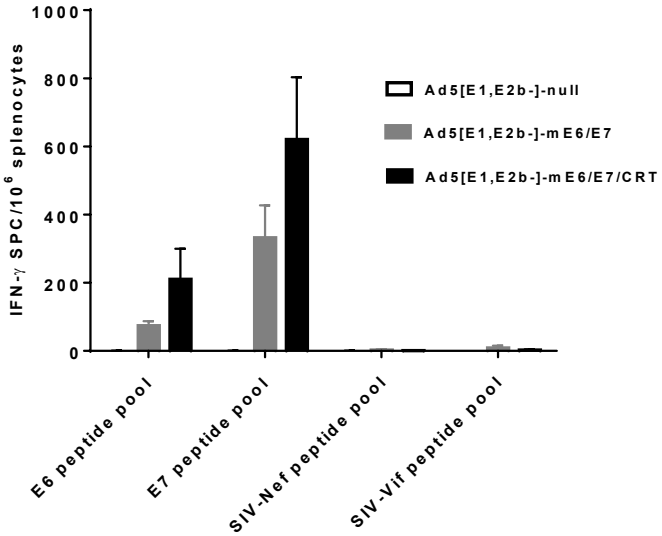


Figure 13. Non-tumor bearing mice were treated as described in **Figure 11** above. Two weeks after the last treatment, mice were assessed for CMI activity as determined by ELISpot assays for IFN-γ secreting splenocytes. Note the increased CMI responses in mice treated with combination therapy (Ad5 [E1-, E2b-]-mE6/E7/CRT).

1.2.5 Anti-PD-L1 Monoclonal Antibody: Avelumab

Programmed death ligand 1 (PD-L1) is a transmembrane protein that was first identified for its role in the maintenance of self-tolerance and prevention of autoimmunity.[34] Engagement of PD-L1 on dendritic cells with the programmed death 1 (PD-1) receptor on T cells delivers an inhibitory signal that promotes T cell anergy or apoptosis.[35] This immunoinhibitory checkpoint is often subverted by tumor cells that over-express PD-L1 in order to escape immunosurveillance in the tumor microenvironment. Indeed, there is a strong correlation between PD-L1 expression and prognosis in cancer. Blockade of the interaction between PD-L1 on tumor cells and PD-1 on T cells is expected to reverse T cell suppression within tumors, thereby promoting effective anti-tumor immune responses.

Several antibodies directed against the PD-L1 / PD-1 pathway are in clinical development for cancer treatment.[36] Compared with anti-PD-1 antibodies that target T-cells, anti-PDL1 antibodies that target tumor cells are expected to have less side effects, including a lower risk of autoimmune-related safety issues, as blockade of PD-L1 leaves the PD-L2 / PD-1 pathway intact to promote peripheral self-tolerance.[37] To this end, avelumab, a fully human IgG1 anti-PDL1 antibody (drug code MSB0010718C) has been produced. Avelumab selectively binds to PD-L1 and competitively blocks its interaction with PD-1.

Avelumab is also cross-reactive with murine PD-L1, thus allowing in vivo pharmacology studies to be conducted in normal laboratory mice. However, due to immunogenicity directed against the fully human avelumab molecule, the dosing regimen was limited to three doses given within a week. The key preclinical pharmacology findings for avelumab are summarized below.

- Functional enhancement of primary T cell activation in vitro in response to antigen-specific and antigen non-specific stimuli.
- Significant inhibition of in vivo tumor growth (PD-L1 expressing MC38 colon carcinoma) as a monotherapy.
- In vivo efficacy is driven by CD8+ T cells, as evidenced by complete abrogation of anti-tumor activity when this cell type was systemically depleted.
- Combination with localized, fractionated radiotherapy resulted in complete regression of established tumors with generation of anti-tumor immune memory.
- Chemotherapy combinations also showed promising activity:
    - Additive combination effect when partnered with oxaliplatin and 5-fluorouracil (5-FU) (core components of FOLFOX [oxaliplatin, 5-FU, and folinic acid]) against MC38 colon tumors. (Summarized in section 2.5)
    - Significant increase in survival when partnered with gemcitabine against PANC02 pancreatic tumors.
- Antibody-dependent cell-mediated cytotoxicity (ADCC) was demonstrated against human tumor cells in vitro; furthermore, studies in ADCC deficient settings in vivo support a contribution of ADCC to anti-tumor efficacy.
- No complement-dependent cytotoxicity was observed in vitro.
- Immunomonitoring assays with translational relevance for the clinic further support an immunological mechanism of action:
    - Consistent increases in CD8+PD-1+ T cells and CD8+ effector memory T cells as measured by fluorescence-activated cell sorter (FACS).

- Enhanced tumor-antigen specific CD8+ T cell responses as measured by pentamer staining and enzyme-linked immunosorbent spot (ELISPOT) assays.

1.2.5.1 Outcomes in colorectal cancer in trials using agents blocking PD-1 – PD-L1 interaction

Despite reports indicating that anti-tumor radiographic responses were unlikely using agents that interfere with PD-1 -- PD-L1 binding in colorectal cancer,[38] there have been reports of radiographic responses.[39-41] Additionally, a correlation has been demonstrated in multiple clinical trials indicating that PD-L1 expression levels on tumor tissue predict the likelihood of radiographic response.[38,40] However, it has become clear that PD-L1 expression, as it is currently measured, is not a definitive requirement for anti-tumor efficacy.[39,40] It has been noted that colorectal tumors rarely express PD-L1 compared with other tumors that are more likely to respond to PD-1 – PD-L1 blockade.[38] However, it is known that a strong anti-tumor T cell response, producing IFN-gamma, will induce PD-L1 expression. Together, this data strongly supports the conclusion that an underlying immune response is necessary for PD-1 – PD-L1 blockade to have an anti-tumor effect. This is part of the rationale for this combination with the standard therapy and Ad-CEA vaccine that may be capable of induction of PD-L1 expression (described in section **1.2.8** below) and thereby increase the anti-tumor activity of PD-1 – PD-L1 blockade.

1.2.5.2 Avelumab Phase I (13-C-0063) Safety

We have recently completed the dose escalation portion of clinical trial 13-C-0063 (NCT01772004). 4, 14 (1 withdrew before treatment), 12 and 21 patients, respectively, were enrolled into 4 escalating dose levels: 1 mg/kg, 3 mg/kg, 10 mg/kg, and 20 mg/kg. The maximum tolerated dose was not exceeded. At the 20 mg/kg dose level, 1 dose limiting toxicity was experienced out of 6 patients treated. This DLT was associated with a significant anti-tumor response and was attributed to drug because of the possibility that it was an immune-related adverse event (with transient elevated creatine kinase, muscle weakness, myalgia, transaminitis, myocarditis with elevated troponin and inferiolateral ST-elevation on ECG). To date, 80 patients have been enrolled. 55pts (71.2%) have come off-study: 43 (53.7%) for progression and 6 (7.5 %) for toxicity. Grade 3 AEs attributable to drug comprised of 7 laboratory abnormalities (4 increased liver transaminases, 1 decreased lymphocytes,1 elevated creatine kinase and 1 elevated lipase) in 5 patients and symptomatic myositis requiring intervention in 2 other patients

1.2.5.3 Pharmacokinetics and Receptor Occupancy data

Data from 25 patients was evaluable for PK and RO analysis. Median time to maximum concentration was approximately 1.5 to 2 hours after infusion (for all doses with a linear PK). Half-life was 63.4, 80.7, 93.9 and 115.1 hours for dose levels 1, 2, 3 and 4 respectively, measured by ELISA. Target RO data was available for 13 patients, measured by PD-L1 binding on peripheral leucocytes via flow cytometry. Mean RO was 75.7, 93.8 and 93.2% for dose levels 1, 2 and 3 respectively.

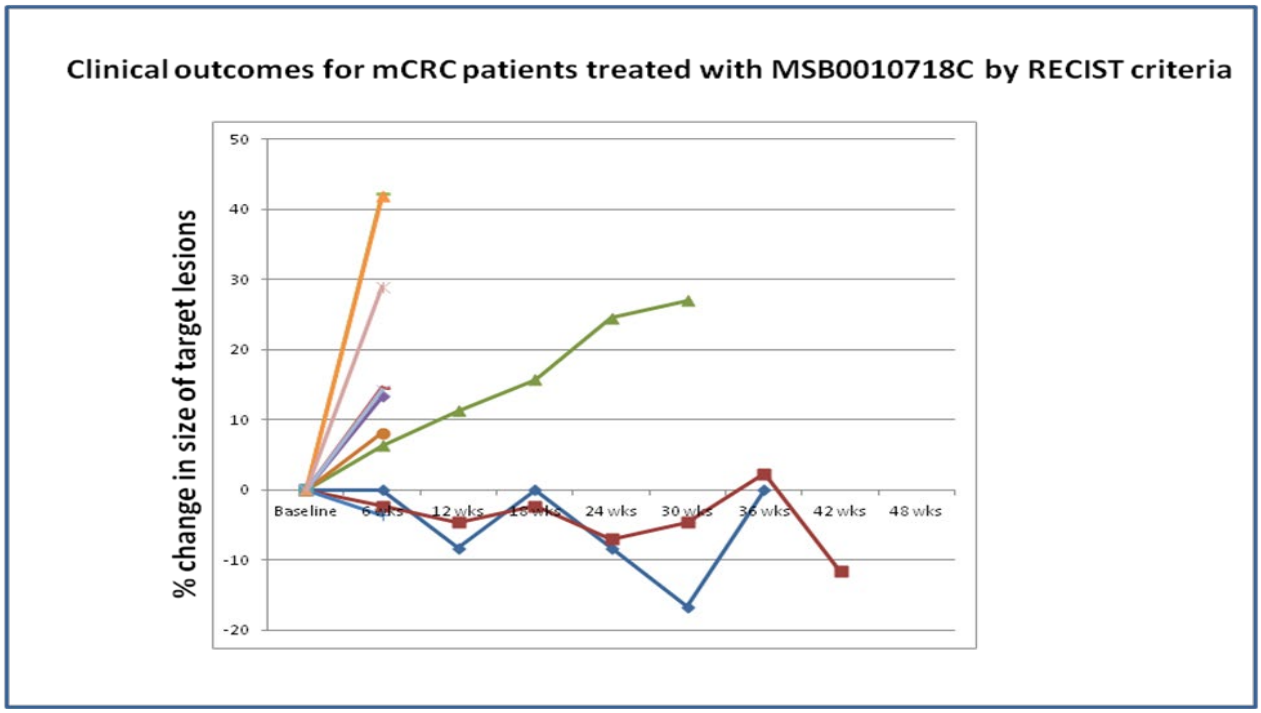
1.2.5.4 Clinical outcomes in patients with colorectal cancer

1.2.5.4.1 Progression Free survival (PFS)

At the time of the writing of this document, 18 patients have enrolled on the single-agent phase I study of avelumab with mCRC at the 10mg/kg dose level. Of these 18, 14 had reached the first restaging for evaluation. Of the 14 evaluable patients, 3 remain on study without evidence of progression for >100 days. Two of these patients have had stable disease for >200 days. The patient

to be detailed in section **1.2.5.4.2** with a significant fall in CEA has remained on study with stable disease through day 350. Of the remaining evaluable patients (n = 11), 9 have had disease progression within 85 days of initial treatment. The remaining 2 patients came off study for adverse events (transaminase elevation and infusion reaction, respectively). **Figure 14** illustrates the outcomes to date for patients with CRC treated with avelumab (MSB0010718C) by Response Evaluation Criteria for Solid Tumors (RECIST) criteria.

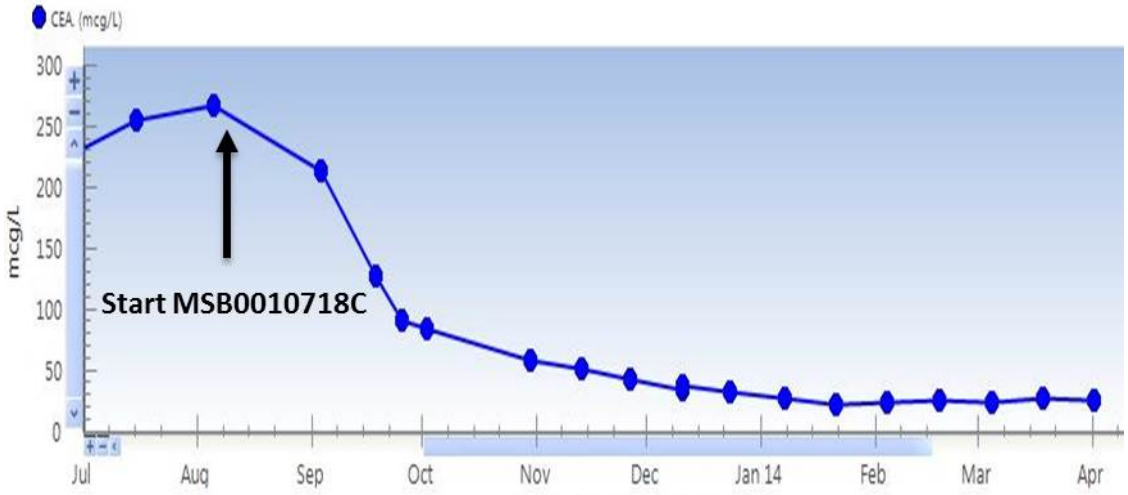
Figure 14. Observed changes in target lesions by size (RECIST)



1.2.5.4.2 CEA response

One patient with mCRC on the phase I study had a remarkable decrease in serum CEA. At baseline his CEA was 267. After starting avelumab, he had a rapid and consistent fall in CEA over the following 5 months with stable disease on scans. He has now been on study for over 11 months with stable disease on scans. Notably, a biopsy of one of his retroperitoneal lymph nodes, being followed as measurable disease on CT scan, revealed "fibroadipose tissue with focal chronic inflammation. No tumor seen." This patient still has elevated CEA in the range of 22-41 that has been holding in this range for 5 months. **Figure 15** demonstrates the change in this patient's CEA.

Figure 15. CEA change in patient with mCRC receiving single agent avelumab (MSB0010718C)



1.2.5.4.3 Immune data

An analysis of immune cell subset changes from baseline to day 15 and day 43 was performed on PBMC samples from the first 23 patients enrolled on the phase I study of avelumab on clinical trial 13-C-0063 (NCT01772004), which included patients at 4 dose levels (1, 3, 10, and 20mg/kg). There was no clear pattern of change (defined as 20% increase or decrease) pre- versus post-treatment in absolute lymphocyte count. A panel of markers was used to define >50 immune cell subsets by flow-cytometry at the same intervals. There were no significant changes in any of the immune cell subsets. Notably, there was no decrease in the number of circulating immune cells expressing PD-L1 (including B lymphocytes and dendritic cells).

1.2.6 Colorectal cancer standard of care

1.2.6.1 FOLFOX (5-fluorouracil, leucovorin, oxaliplatin)

A randomized trial comparing irinotecan and bolus fluorouracil plus leucovorin (IFL, control combination), oxaliplatin and infused fluorouracil plus leucovorin (FOLFOX), or irinotecan and oxaliplatin (IROX) established the FOLFOX combination, given for a total of 6 months, as the standard of care for first line treatment in patients with metastatic colorectal cancer (mCRC).[42] Though multiple infusion schedules of FOLFOX have been validated, typically denominated as 'modified FOLFOX, there are no essential changes in the constituent cytotoxic agents of the regimen. Of these, mFOLFOX6 is one of the most widely used.

Oxaliplatin, however, is very difficult for patients to receive for greater than 6 months (12 cycles) due to progressive neurotoxicity. Though 6 months of combination therapy remains the standard of care in mCRC, clinical judgment may influence the decision to limit the number of oxaliplatin-containing cycles towards the end of treatment Other trials, including the CAIRO3 study, have demonstrated the feasibility and benefit of discontinuation of oxaliplatin after a 3 month "induction" period with continuation of 5-FU and leucovorin as "maintenance" therapy.[43]

1.2.6.2 Bevacizumab (Avastin®)

Addition of bevacizumab to first-line 5-FU and Oxaliplatin containing regimens was demonstrated to increase time to progression in mCRC patients with a manageable side effect profile and non-overlapping toxicities.[44,45] Later trials indicated that continuing bevacizumab beyond first progression (in combination with subsequent chemotherapy) improved overall survival in an unselected group of patients by KRAS mutational status, which has led to its approved use in the maintenance setting.[46,47]

1.2.6.3 Capecitabine

This agent is a prodrug that is enzymatically converted to 5-fluorouracil by 3 enzymatic steps following oral ingestion. As an orally active fluoropyrimidine, capecitabine has been approved for use in the adjuvant setting.[48] In the advanced colon cancer setting, it has been shown to be equally efficacious as 5-fluorouracil, though with more reported rates of hand-foot syndrome.[49] This agent offers the convenience of the oral route with its benefits of reducing infusion commitments for patients in the maintenance setting, while achieving high concentrations intratumorally, given the higher concentrations of thymidine phosphorylase in tumor as compared to normal tissues.[50]

1.2.6.4 Population expected outcomes

Using the above noted standard of care, the expected radiographic progression free survival is approximately 10 months in patients with metastatic colorectal adenocarcinoma.[46,47] At the time of progression, patients will typically be treated with a similar regimen, using 5-FU, leucovorin, and irinotecan (FOLFIRI). Unfortunately, that therapy also has a limited duration of benefit and patients have very few choices in the third line setting. Clearly, there is a significant need to improve outcomes in patients overall with metastatic colorectal cancer. Combining agents with non-overlapping toxicity and different mechanisms of action in the front line setting may be an excellent way to improve short and long term clinical outcomes.

1.2.7 Rationale for combination of FOLFOX-A, Ad-CEA vaccine, and Avelumab

As previously noted, PD-1/L1 blockade has had minimal effect in the majority of patients with advanced colorectal cancer. The notable exception has been patients with microsatalite instability (MSI-Hi, also called MisMatch Repair Deficiency, MMR-D).[51] It has been hypothesized that the increased mutation burden in these tumors provides a greater number of potential neo-epitopes for T-cell receptors to recognize and bind, resulting in killing.[52,53] Combining anti-PD-L1 antibody with a therapeutic cancer vaccine capable of inducing CEA-specific T cell immune responses plus the standard of care creates multiple advantages that may allow for improved clinical outcomes.

As previously discussed, advanced colorectal cancer appears to be weakly immunogenic. Therapeutic cancer vaccines are designed to drive T cell activation against a tumor-associated antigen. The Ad-CEA vaccine targets CEA, an antigen overexpressed universally in colorectal cancer. T cell activation against the target antigen should result in greater trafficking and infiltration of T cells in to the tumor microenvironment. When combined with Avelumab, a PD-L1 inhibitor, those activated T cells are more likely to have a direct anti-tumor effect, resulting in radiographic tumor response and delayed time to progression.

Both 5-FU and oxaliplatin have been identified as agents which can induce immunogenic cell death and/or decrease the number of regulatory T cells [54], which increases the antigen targets

available to active T cells and T cell activation and killing of tumor cells. This effect is known to be capable of inducing an anti-tumor immune response.[54-58] There is also evidence that immunotherapy in combination with 5-FU is capable of increasing the immune response against tumor cells. As has been discussed, an underlying anti-tumor T cell response appears to be necessary for PD-L1 induction in the tumor tissue. PD-L1 expression has been correlated with the likelihood of PD-1 – PD-L1 blockade to induce anti-tumor immune response and cause tumor volume reduction and clinical benefit.

Another mechanism by which the underlying anti-tumor immune response may be improved when patients are exposed to the standard of care regimen is through a process known as immunogenic modulation. Hodge and colleagues in the LTIB have demonstrated repeatedly the potential improvement in antigen presentation, FAS upregulation, and T cell mediated killing in combination with chemotherapeutics, including 5-FU and oxaliplatin.[59]

Decreased tumor burden induced by standard of care therapy may allow better immune response by debulking the tumor and improving the microenvironment for immunologic killing.[60-65] The addition of standard of care therapy also provides ample time for an immune response to occur, which may be important considering the occasional delayed responses and prolonged stable disease findings that have been described with anti-PD-1 and anti-PD-L1 targeting agents. Given the findings with ipilimumab improving overall survival while not necessarily improving PFS[66], it may be that delayed immunologic effects create long term clinical benefit in some populations despite lack of immediate tumor volume reduction. By using a therapy in combination with standard agents known to induce tumor volume reduction, we may be able to capture that long term benefit.

Preclinical evidence indicates that vascular endothelial growth factor (VEGF) signaling in the tumor microenvironment causes disorganized perfusion, resulting in poor T cell trafficking to the tumor site, increased hypoxia, and an immunosuppressive microenvironment. Rakesh Jain's group has demonstrated significant improvement in T cell infiltration with the addition of VEGF inhibition in preclinical models.[67] In this study, VEGF inhibition increased the efficacy of a vaccine to induce T cell infiltration into the tumor, resulting in better anti-tumor effect. Similarly, Dr. Hwu's group has found VEFG blockade to result in greater T cell infiltration in adoptively transferred T cells in a murine model.[68] Alfaro, et al., found VEGF inhibition to be valuable for dendtritic cell differentiation and T cell activation.[69] Taken together, these data indicate a potential synergy between VEGF inhibition and immunotherapy, supportive of combining these agents in patients.

1.2.7.1 Rationale for exclusion of MSI-high mCRC

While PD-1/L1 blockade has resulted in objective tumor responses in patients with microsatellite instability, there is an unmet need for better therapeutic interventions for patients who have microsatellite stable colorectal cancer. In these patients, who make up the large majority of patients with metastatic colorectal cancer (>90%), there has been little or no evidence of clinical benefit with PD-1/L1 blockade. Our hypothesis is that the combination of the agents comprising FOLFOX-bevacizumab and Ad-CEA vaccine will increase specific T cell activation, which will result in PD-1/L1 blockade allowing T cell mediated killing of tumor cells. As a result, patients who are known to respond to PD-1/L1 blockade are excluded from this study as they may not need chemotherapy at all. Ongoing studies in the population of MSI-Hi colorectal cancer are

ongoing to answer this question and we would not want to confound our study by including patients in the MSI-Hi population.

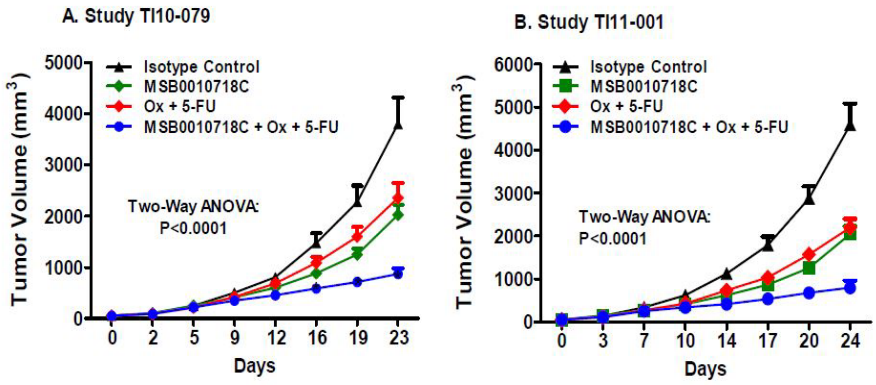
1.2.7.2 Preclinical data support

Our CRADA partner, EMD-Serono, has already performed a preclinical model evaluation of the combination of FOLFOX with anti-PD-L1. The study was conducted in compliance with GLP and the global policy "Quality management system (QMS) for Merck Serono research (MSR)" of Merck KGaA. Materials and methods and additional data available in **Appendix A**.

The study investigated the potential of combining avelumab with the core components of FOLFOX (5-fluorouracil and oxaliplatin) in two separate studies (TI10-079 & TI11-001) conducted in mice bearing subcutaneous MC38 tumors.

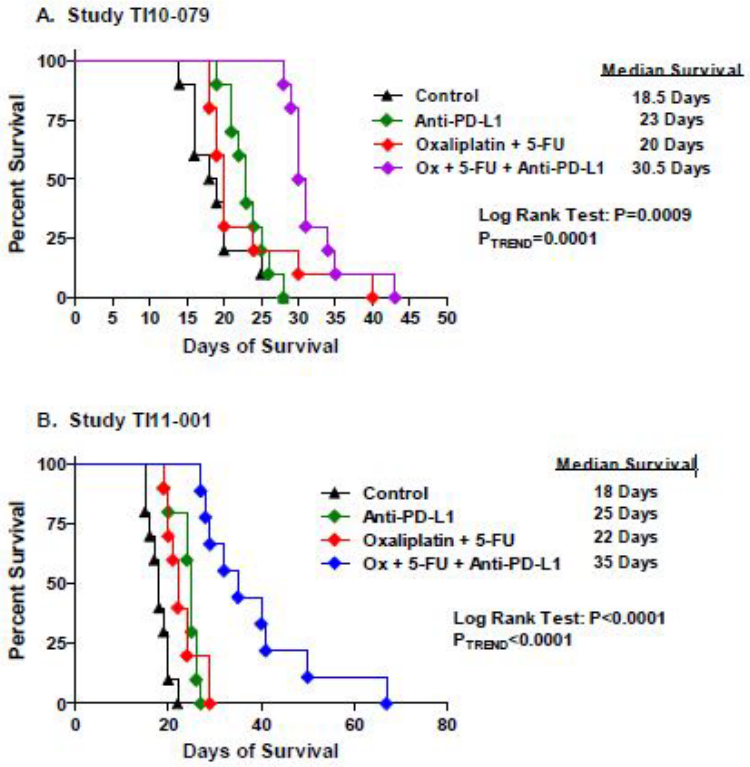
The therapeutic combination of oxaliplatin/5-FU and anti-PD-L1 blockade using avelumab in MC38 tumor-bearing mice yielded superior tumor growth inhibition relative to control mice and either form of monotherapy (**Figure 16**). Additionally, the avelumab and oxaliplatin/5-FU combination significantly extended survival compared to mice receiving either vehicle control or monotherapy (**Figure 17**).

Figure 16. MC 38 Tumor Growth Inhibition in Mice Receiving Avelumab (Anti-PD-L1) in Combination with Oxaliplatin and 5-FU



Female C57BL/6 mice were inoculated in the right subcutaneous flank with 1x106 MC38 colon carcinoma cells. When tumors reached a mean volume of ~50 mm3, mice were sorted into treatment groups (N=10). 5-FU (60 mg/kg by i.v. injection) and OX (5 mg/kg by i.p. injection) were administered on days 0 and 14. MSB0010718C (400 µg by i.v. injection) was given on days 3, 6, and 9. Tumor volumes were measured twice weekly via calipers. Data are mean ± SEM.

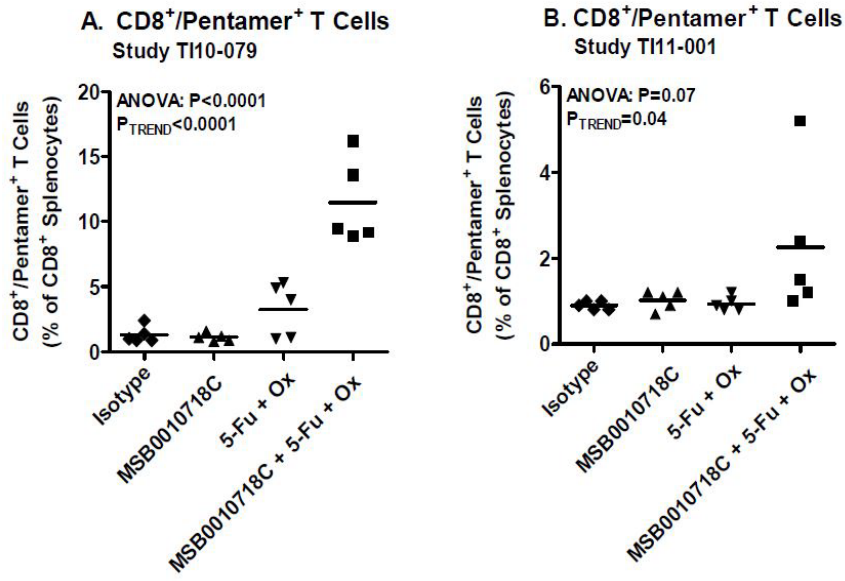
Figure 17. Survival of MC38 Tumor-Bearing Mice Receiving Avelumab (Anti-PD-L1) in Combination with Oxaliplatin and 5-FU



Female C57BL/6 mice were inoculated in the right subcutaneous flank with $1x10^6$ MC38 colon carcinoma cells. When tumors reached a mean volume of ~50 mm3, mice were sorted into treatment groups (N=10). 5-FU (60 mg/kg by i.v. injection) and OX (5 mg/kg by i.p. injection) were administered on days 0 and 14. MSB0010718C (400 µg by i.v. injection) was given on days 3, 6, and 9. Kaplan-Meier plots and Log Rank statistics were used to calculate survival differences between groups. Survival was censored at the time tumor volumes reached 2000 mm3. Data are expressed as median survival time in days.

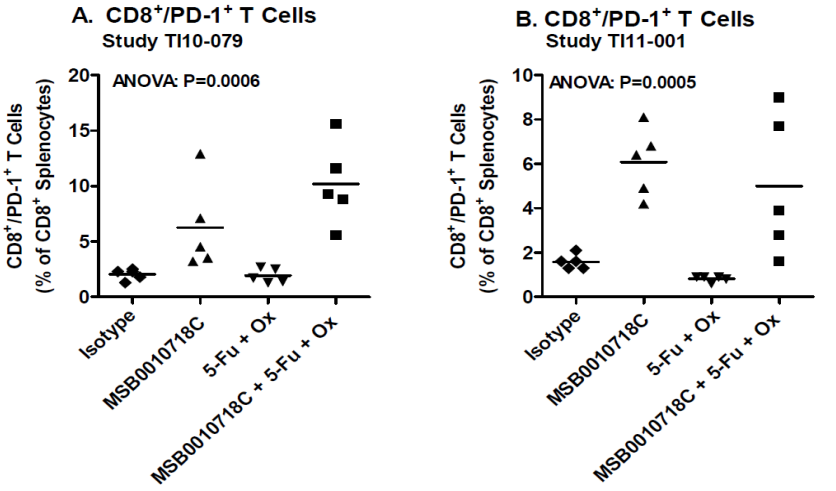
In both *in vivo* studies (TI10-079 and TI11-001), a subset of mice in each treatment group was sacrificed to evaluate trends in immune phenotype in the spleen and tumor tissues. Using a pentamer analysis of splenic tissue, the avelumab and oxaliplatin/5-FU combination was shown to significantly elevate the precursor frequency of tumor-reactive P15E-specific CD8+ T cells (P15E is a known tumor antigen expressed in MC38 colorectal cancer tumor lines,[70] (**Figure 18**). Additionally, avelumab therapy was shown to consistently and significantly increase the percentage of splenic CD8+ T cells expressing the PD-1 receptor (**Figure 19**).

Figure 18. Pentamer Analysis of Tumor-Antigen Specific T Cells



Subsets of 5 animals from each study group were sacrificed on study day 21 for FACS-based detection of p15Especific T cells using a fluorescently-labeled synthetic MHC class I pentamer loaded with a p15E-derived antigenic peptide epitope. The spleen from each animal was dissociated into single cell suspensions and subsequently stained with an anti-CD8 antibody and the p15E pentamer. From the total splenocyte gate, the percentage of cells positive for both CD8 and the pentamer was determined. Data were acquired using a BD LSR II flow cytometer.

Figure 19. Splenic CD8+/PD-1+ T cell Counts

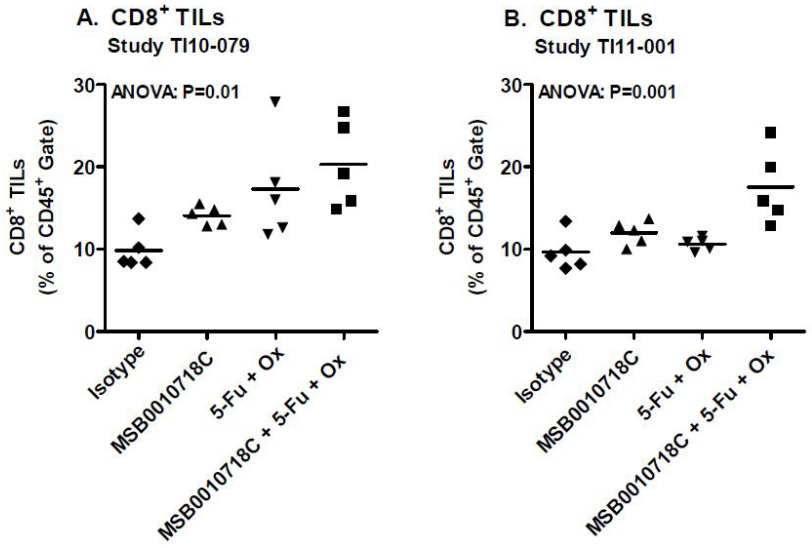


Subsets of 5 animals from each group were sacrificed for FACS-based immunophenotyping on day 21 after the start of treatment. Spleens were harvested and dissociated into single cell suspensions by mechanical disruption.

Suspensions were then stained with fluorescently-labeled antibodies specific for relevant T cell surface markers (i.e. CD8, PD-1, CD44, CD62L) and data were acquired on a BD LSR II flow cytometer.

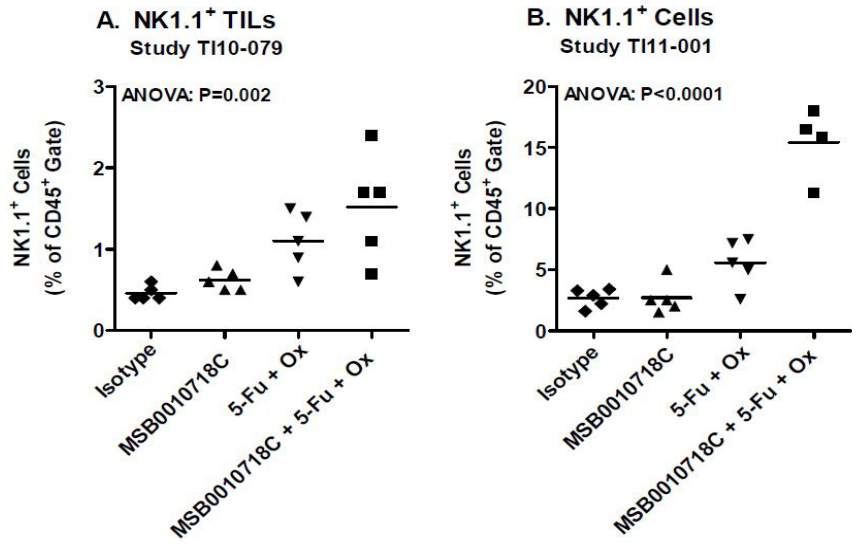
Consistent with our findings in the spleen, intratumoral CD8+ T cells and NK1.1+ cells were elevated in mice receiving combination therapy with avelumab and oxaliplatin/5-FU (see **Figure 20** and **Figure 21**, respectively). Taken together, the phenotypic data from mice treated with the avelumab and oxaliplatin/5-FU combination suggest that this immunotherapeutic regimen is successfully activating immune effector cell populations with known tumoricidal potential (NK1.1+ and CD8+ T cells).

Figure 20. CD8+ Tumor-Infiltrating Lymphocyte Levels



Subsets of 5 animals from each group were sacrificed for FACS-based immunophenotyping analysis on day 21 following the start of treatment. Tumors were harvested and dissociated into single cell suspensions by enzymatic digestion (collagenase) and mechanical disruption. Suspensions were then stained with a fluorescently-labeled antibodies specific for the NK1.1 surface marker and data were acquired using a BD LSR II flow cytometer.

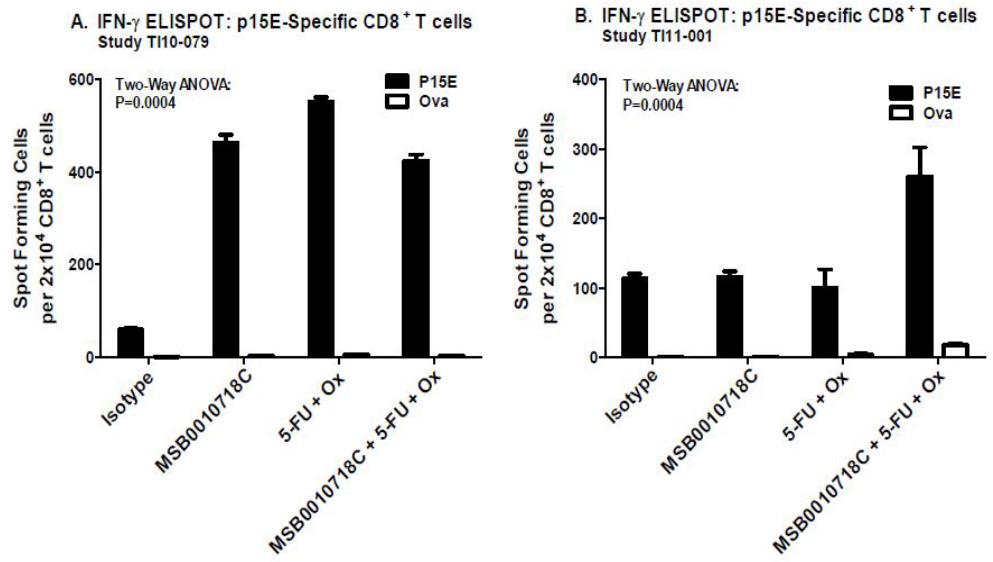
Figure 21. Tumor-Infiltrating NK Cell Levels



Subsets of 5 animals from each group were sacrificed for FACS-based immunophenotyping analysis on day 21 following the start of treatment. Tumors were harvested and dissociated into single cell suspensions by enzymatic digestion (collagenase) and mechanical disruption. Suspensions were then stained with a fluorescently-labeled antibodies specific for the NK1.1 surface marker and data were acquired using a BD LSR II flow cytometer.

The frequency of IFN-γ producing CD8+ T effector cells directed against the p15E tumor antigen was also measured using an ELISPOT assay that detects IFN-γ secretion as a marker of T cell activation. Using the ELISPOT assay, a strong p15E-specific response was detected in all treated groups in study TI10-079 (see **Figure 22A**). In study TI11-001, the highest p15E-specific response was measured in the combination treatment group (see **Figure 21B**).

Figure 22. Combination of Avelumab (MSB0010718C) with 5-FU and Oxaliplatin: ELISPOT Measurement of Tumor Antigen Specific CD8+ T cells



For the ELISPOT assay, spleens from treated animals were harvested on day 21 after the start of treatment and processed into single cells suspensions (splenocytes from 5 mice from each group were pooled). The splenocytes were cultured for six days in the presence of a p15E-derived peptide antigen epitope (amino acid sequence KSPWFTTL) in order to stimulate expansion of antigen-specific CD8+ T cells. As a control for non-specific activation, the assay was run in parallel using an irrelevant peptide derived from chicken ovalbumin (OVA). Following the six day period of *in vitro* antigen stimulation, the CD8+ T cells were isolated from the cultures and placed in co-culture with antigen presenting cells (APCs) loaded with the same p15E antigenic peptide or OVA as a control. The cocultures were prepared in ELISPOT assay plates coated with an antibody that captured secreted IFN-γ. Following assay plate development, each IFN-γ positive "spot" represented a CD8+ T cell that had become activated by p15E-specific stimulation.

One interpretation of this data is that chemotherapy initiates a cascading sequence in which tumor cell destruction and the shedding of tumor antigen in the tumor microenvironment activates innate and adaptive immune responses to the tumor. In this scenario, PD-L1 blockade is thought to potentiate the adaptive immune response, which may indirectly activate innate effector cells such as NK cells.

By blocking negative costimulation of tumor-reactive T cells mediated by the PD-1/PD-L1 interaction, avelumab may be employed in a variety of settings and indications to maximize the potential efficacy of standard of care cytotoxic therapies as well as immunotherapy combinations.

1.2.7.3 Clinical data support

We have looked into the use of standard chemotherapy in patients with previously untreated metastatic colorectal cancer and have found no difference in the number of 40+ different immune cell subsets pre- versus post-treatment with standard therapy (5-FU based chemotherapy[71]). This was done in collaboration with Dr. John Marshall at Georgetown, who assisted with collecting

clinical samples from 6 patients pre- and post-standard therapy. Thus, chemotherapy should not diminish the potential immune benefits of anti-PD-L1 therapy.

1.2.7.4 Clinical safety data combining FOLFOX + PD-1/PD-L1+ Bevacizumab therapy

The first patient enrolled on this trial developed febrile neutropenia and shortly thereafter passed away from a grade 5 bowel perforation event attributed to bevacizumab. Data from a phase II trial of a mFOLFOX6 + bevacizumab + a PD-L1 inhibitor [72] showed no reported episodes of bowel perforation among 30 patients with colorectal cancer enrolled on this combination. Given this data and the fact that the episode of bowel perforation on this study is a solo event, we think it is appropriate to continue with bevacizumab on the combined chemotherapy/ immunotherapy arm (Arm B) of the study for now. Should any similar events of bowel perforation occur we will plan on discontinuing bevacizumab from this arm of the study.

In addition, in the phase II trial of mFOLFOX6 + bevacizumab + a PD-L1 inhibitor cited above there was an unexpectedly high percentage of grade ≥ 3 neutropenia (43%) in patients receiving this combination. In addition, a second phase II trial [73] evaluating mFOLFOX6 + pembrolizumab had an unexpectedly high rate of febrile neutropenia (3/6 pts) in their safety lead in cohort. As it is generally standard clinical practice to first remove the 5-FU bolus infusion in the setting of neutropenia and based upon the expected high rate of neutropenia with this combination, we are prophylactically removing the 5-FU bolus infusion from the chemotherapy/ immunotherapy arm (Arm B) of the study.

1.2.8 Summary

- Colorectal cancer (CRC) is the fourth most common cancer diagnosis in the United States and accounts for the second most cancer-related deaths.
- Programmed death ligand 1 (PD-L1) is a transmembrane protein that was first identified for its role in the maintenance of self-tolerance and prevention of autoimmunity. Blockade of the interaction between PD-L1 on tumor cells and PD-1 on T cells is expected to reverse T cell suppression within tumors. These agents are dependent on underlying T cell activation against the tumor cell to be effective.
- Avelumab is a fully human IgG1 anti-PDL1 antibody that selectively binds to PD-L1 and competitively blocks its interaction with PD-1.
- In ongoing phase 1 trials of avelumab, the agent has been well tolerated and has shown clinical activity.
- Clinical trials with anti-PD-1/L1 agents in colorectal cancer have resulted in minimal activity in patients who do not have mismatch repair deficiency (MMR-D)
- Therapeutic cancer vaccines targeting overexpressed proteins offer a potential method to activate T cells against tumors.
- A novel adenovirus based, CEA-targeting vaccine has demonstrated cytolytic T cell responses in patients with metastatic colorectal cancer.
- Standard of care agents in first line metastatic CRC have properties been associated with improved immune response via immunologic cell death and immunogenic modulation
- A preclinical model combining 5-FU and oxaliplatin plus avelumab has demonstrated potential synergy of this combination

2 ELIGIBILITY ASSESSMENT AND ENROLLMENT

2.1 ELIGIBILITY CRITERIA

2.1.1 Inclusion Criteria

2.1.1.1 Subjects must have previously untreated metastatic or unresectable colorectal cancer and have no contraindications to treatment with the standard of care regimen (as defined in section **3.4.1**) as determined by the investigator. Prior adjuvant therapy for surgically resectable disease (including oligometastatic disease) is acceptable (including immunotherapy), but must have been completed at least 6 months prior to enrollment.

2.1.1.2 Patients should not be eligible for potentially curative surgical intervention in the case of oligometastic disease at the time of enrollment or must have actively refused after explicit discussion of potential benefit of this intervention with multidisciplinary team.

2.1.1.3 Histologically confirmed colorectal cancer.

2.1.1.4 Patients must have measurable disease by RECIST criteria as defined in section **6.4**.

2.1.1.5 Age ≥18 years. Because safety data is not known with this agent in patients less than 18 years old, children are excluded from this study.

2.1.1.6 ECOG performance status ≤ 2 (see **Appendix B**).

2.1.1.7 Patients must have normal organ and marrow function as defined below:

- Creatinine clearance (Per Institutional standard or 24-hour urine) ≥ 30 mL/min.
- Adequate hepatic function defined by a total bilirubin level ≤ 1.5 × the upper limit of normal range (ULN), an aspartate aminotransferase (AST), level ≤ 2.5 × ULN, and an alanine aminotransferase (ALT) level ≤ 2.5 × ULN or, for subjects with documented metastatic disease to the liver, AST and ALT levels ≤ 5 × ULN
- Hematological eligibility parameters (within16 days of enrollment):
    - Granulocyte count ≥ 1,500/mm^3
    - Platelet count ≥100,000/mm^3
    - Hemoglobin ≥ 9 g/dL

2.1.1.8 The effects of Ad-CEA vaccine and avelumab on the developing human fetus are unknown. For this reason and because Ad-CEA vaccine and avelumab as well as other therapeutic agents used in this trial are known to be teratogenic, women of child-bearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry and for the duration of study participation and for a period of 4 months after the last treatment with avelumab or 6 months after the last administration of bevacizumab, whichever occurs later. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately.

2.1.1.9 Ability of subject to understand and the willingness to sign a written informed consent document.

2.1.2 Exclusion Criteria

2.1.2.1 Metastatic or unresectable colorectal cancer with mismatch repair deficiency (MMR-D or MSI-High).

2.1.2.2 Concurrent treatment for cancer except agents specified within the treatment protocol.

2.1.2.3 Prior major surgery or gastrointestinal perforation within 28 days of enrollment.

2.1.2.4 Persisting toxicity related to prior therapy (NCI CTCAE v5.0 Grade > 1); however alopecia, sensory neuropathy Grade <=2, or other Grade <=2 AEs not constituting a safety risk based on investigator's judgment are acceptable.

2.1.2.5 Known history of testing positive for HIV or known acquired immunodeficiency syndrome.

2.1.2.6 Hepatitis B virus (HBV) or hepatitis C virus (HCV) infection at screening (positive HBV surface antigen or HCV RNA if anti-HCV antibody screening test positive).

2.1.2.7 Any significant disease that, in the opinion of the investigator, may impair the patient's tolerance of study treatment.

2.1.2.8 Active autoimmune disease that might deteriorate when receiving an immuno-stimulatory agent. Patients with diabetes type I, vitiligo, psoriasis, or hypo- or hyperthyroid diseases not requiring immunosuppressive treatment are eligible.

2.1.2.9 Current use of immunosuppressive medication, EXCEPT for the following: a. intranasal, inhaled, topical steroids, or local steroid injection (e.g., intra-articular injection); b. Systemic corticosteroids at physiologic doses ≤ 10 mg/day of prednisone or equivalent; c. Steroids as premedication for hypersensitivity reactions (e.g., CT scan premedication).

2.1.2.10 Patients who are receiving any other investigational agents within 28 days before start of study treatment.

2.1.2.11 Prior organ transplantation including allogenic stem-cell transplantation.

2.1.2.12 Subjects with active central nervous system (CNS) metastases causing clinical symptoms or metastases that require therapeutic intervention are excluded. Subjects with a history of treated CNS metastases (by surgery or radiation therapy) are not eligible unless they have fully recovered from treatment, demonstrated no progression for at least 2 months, and do not require continued steroid therapy. Subjects with CNS metastases incidentally detected during Screening which do not cause clinical symptoms and for which standard of care suggests no therapeutic intervention is needed are eligible.

2.1.2.13 Active infection, requiring systemic therapy.

2.1.2.14 Clinically significant (i.e., active) cardiovascular disease: cerebral vascular accident/stroke (< 3 months prior to enrollment), myocardial infarction (< 3 months prior to enrollment), unstable angina, congestive heart failure (≥ New York Heart Association Classification Class II), or uncontrolled arrhythmias.

2.1.2.15 Other severe acute or chronic medical conditions including immune colitis, inflammatory bowel disease, immune pneumonitis, pulmonary fibrosis or psychiatric conditions including recent (within the past year) or active suicidal ideation or behavior; or laboratory abnormalities that may increase the risk associated with study

participation or study treatment administration or may interfere with the interpretation of study results and, in the judgment of the investigator, would make the patient inappropriate for entry into this study.

2.1.2.16 Pregnant women and breastfeeding mothers are excluded due to unknown impact on embryos or infants.

2.1.2.17 Known alcohol or drug abuse.

2.1.2.18 "Known prior severe hypersensitivity to investigational product or any component in its formulations, including known severe hypersensitivity reactions to monoclonal antibodies (NCI CTCAE v5.0 Grade ≥ 3).

2.1.2.19 Patients with a known hypersensitivity/allergy to any of the standard of care agents used in this study or related compounds (e.g. platinum compounds) are excluded.

2.1.2.20 Prior history of hypertensive emergency or hypertensive encephalopathy (for those expected to receive bevacizumab).

2.1.2.21 Serious, non-healing wound, active ulcer, or untreated bone fracture, including tumor-related pathological fracture.

2.1.2.22 Evidence of bleeding diathesis or significant coagulopathy (in the absence of therapeutic anticoagulation).

2.1.2.23 Patients being treated with medications with drug-drug interactions with study agents will require evaluation by to determine if full doses of all study treatments can be given safely. Significant drug-drug interactions will need to be addressed prior to enrollment. Alternatively, the patient will not be eligible.

2.1.2.24 Vaccination within 4 weeks of the first dose of avelumab and while on trials is prohibited except for administration of inactivated vaccines.

2.1.3 Recruitment Strategies

2.2 THIS STUDY WILL BE POSTED ON WWW.CLINICALTRIALS.GOV, NIH WEBSITES AND ON NIH SOCIAL MEDIA FORUMS. SCREENING EVALUATION

2.2.1 Screening activities performed prior to obtaining informed consent

Minimal risk activities that may be performed before the subject has signed a consent include the following:

- Email, written, in person or telephone communications with prospective subjects
- Review of existing medical records to include H&P, laboratory studies, etc.
- Review of existing MRI, x-ray, or CT images
- Review of existing photographs or videos
- Review of existing pathology specimens/reports from a specimen obtained for diagnostic purposes

A waiver of consent for these activities has been requested in section **12.5.2**

2.2.2 Screening activities performed after a consent for screening has been signed

The following activities will be performed only after the subject has signed the consent for this study for screening. Assessments performed at outside facilities or on another NIH protocol within the timeframes below may also be used to determine eligibility once a patient has signed the consent.

Studies should be done within 16 days prior to treatment unless otherwise noted below.

- History and physical exam including performance status and vital signs
- Scans (within 28 days prior to treatment)
  - CT CAP (chest abdomen and pelvis) with oral and IV contrast (MRI abdomen/pelvis with contrast + chest CT without contrast if IV contrast is contraindicated or CT CAP is inadequate)
  - Brain MRI – if clinically indicated
- Hepatic Panel (Alkaline Phosphatase, ALT/GPT, AST/GOT, Total Bilirubin, Direct Bilirubin)
- Acute Care Panel (Sodium [NA], Potassium [K], Chloride [CL] Total CO2 [Bicarbonate], Creatinine, Glucose, Urea nitrogen, eGFR)
- CBC with differential
- Serum or urine HCG in women of childbearing potential
- HBV (HBsAg), HCV (anti-HCV), HIV (anti-HIV) screening (within 3 months prior to treatment)
- Urine dipstick
- PT/INR, PTT
- Histologic confirmation of diagnosis (including MSI or MMR status; any time prior to treatment)

2.3 REGISTRATION PROCEDURES

2.3.1 Registration at the Clinical Center

Registration will be a two-part process as patients are screened on this protocol. Authorized staff must register an eligible candidate with NCI Central Registration Office (CRO) within 24 hours of signing consent. To initially register a subject after the participant has signed the consent, complete the top portion of the registration Eligibility Checklist from the website (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) indicating that the patient is being registered for screening and send via encrypted email to: NCI Central Registration Office ncicentralregistration-l@mail.nih.gov. Once eligibility is confirmed after completion of screening studies, complete the remainder of the form which is the eligibility checklist, indicating that the patient is being registered for treatment and email the completed registration checklist to the CRO at NCI Central Registration Office ncicentralregistration-

l@mail.nih.gov. After confirmation of eligibility at Central Registration Office, CRO staff will call pharmacy to advise them of the acceptance of the patient on the protocol prior to the release of any investigational agents. Verification of Registration will be forwarded electronically via e-mail to the research team. A recorder is available during non-working hours.

Subjects that do not meet screening criteria should be removed from the study following the procedure in section **3.7.2**.

2.3.2 For Participating Site Registration

Registration will be a two-part process as patients are screened on this protocol. A protocol registration form will be supplied by the CCR study coordinator and updates will be provided as needed. To initially register a subject, after the participant has signed consent, complete the top portion of the form and send to CCR study coordinator. Once eligibility is confirmed, after completion of screening studies, complete the remainder of the form which is the eligibility checklist, indicating that the patient is being registered for treatment and send to CCR study coordinator. In addition, source documents supporting the eligibility criteria must be sent to the CCR study coordinator. The CCR study coordinator will notify you either by e-mail or fax that the protocol registration form has been received which will include the unique patient/subject ID number. Questions about eligibility should be directed to the CCR study coordinator or PI. Questions related to registration should be directed to the CCR study coordinator.

Subjects that do not meet screening criteria should be removed from the study following the procedure in section **3.7.4**.

2.3.3 Treatment Assignment and Randomization Procedures

Cohorts

Number	Name	Description
1	Lead in, Cohort 1	Patients with Previously Untreated Metastatic or Unresectable Colorectal Cancer enrolled for safety assessment
2	Cohort 2	Patients with Previously Untreated Metastatic or Unresectable Colorectal Cancer enrolled for efficacy assessment

Arms

Number	Name	Description
A	FOLFOX-A alone	Subjects will receive FOLFOX + bevacizumab for up to 12 2-week cycles followed by maintenance therapy with bevacizumab + capecitabine/5-FU until disease progression.
B	Lead in, FOLFOX-A + Avelumab +Ad-CEA	Subjects will receive FOLFOX + bevacizumab + avelumab + Ad-CEA vaccine (given weeks 0, 2, 4, 8, 12, 16, and then every 12 weeks) for up to 12 2-week cycles followed by maintenance therapy with bevacizumab + capecitabine/5-FU + avelumab + Ad-CEA vaccine (following every 12 weeks dosing schedule per schema in 3.1.1.2) until disease progression.

Randomization and Arm Assignment

Randomization will be performed at study entry by the NCI Central Registration Office. The first 6 evaluable subjects (Cohort 1) will be assigned to the Arm B regimen. Thereafter, if the study proceeds, subjects in Cohort 2 will be randomized on a 1:1 basis to Arm A and Arm B.

3 STUDY IMPLEMENTATION

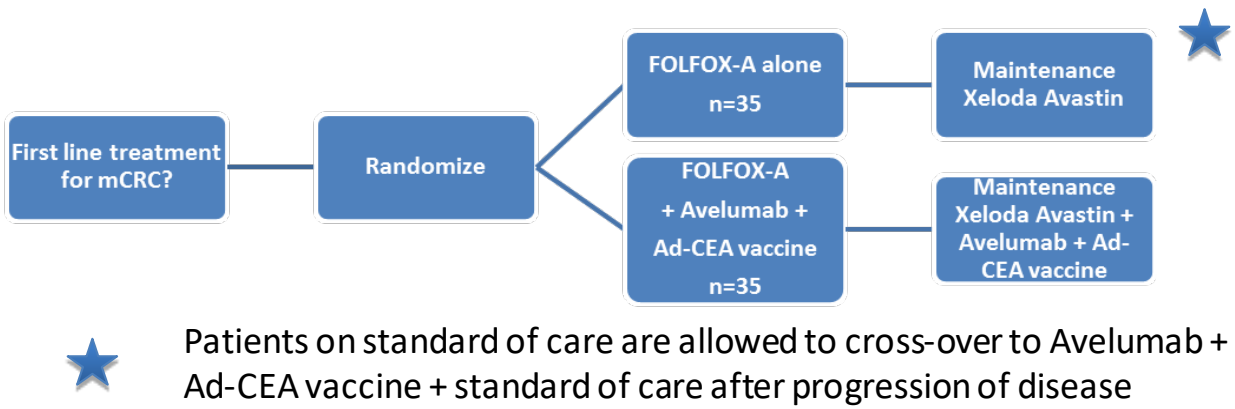
3.1 STUDY DESIGN

This is an open-label, randomized, multicenter phase 2.5 clinical trial designed to evaluate the potential improvement in progression free survival (PFS) when avelumab is used in combination with standard of care therapy and Ad-CEA vaccine in metastatic or unresectable colorectal cancer when compared with standard of care alone. Patients in the standard of care alone arm will be offered the opportunity to cross-over and receive avelumab + Ad-CEA vaccine in combination with any combination of the Arm A standard of care deemed appropriate by the investigator after disease progression (as defined by RECIST 1.1. in section **6.4**). Cross-over will not be offered in the case of rapid or symptomatic progression due to the need for implementation of a standard option with known efficacy. Other clinical and immunologic endpoints will be captured with the goal of establishing correlative data that can later be prospectively tested in a larger phase 3 study.

Initially, we will enroll 6 patients assigned to receive the combination of chemotherapy, Ad-CEA vaccine and avelumab to evaluate safety and feasibility of this treatment schedule. These patients will be enrolled at NCI only. Patients will be enrolled in a staggered fashion with 5 days minimum between each patient enrolled. If there are no dose limiting toxicities, as defined in section **3.2**, we will proceed to the phase 2.5 portion of the trial. If > 1/6 patients experience a DLT attributable to the IND agents, we will not proceed with the phase 2.5 portion of the trial.

3.1.1 Study Schema

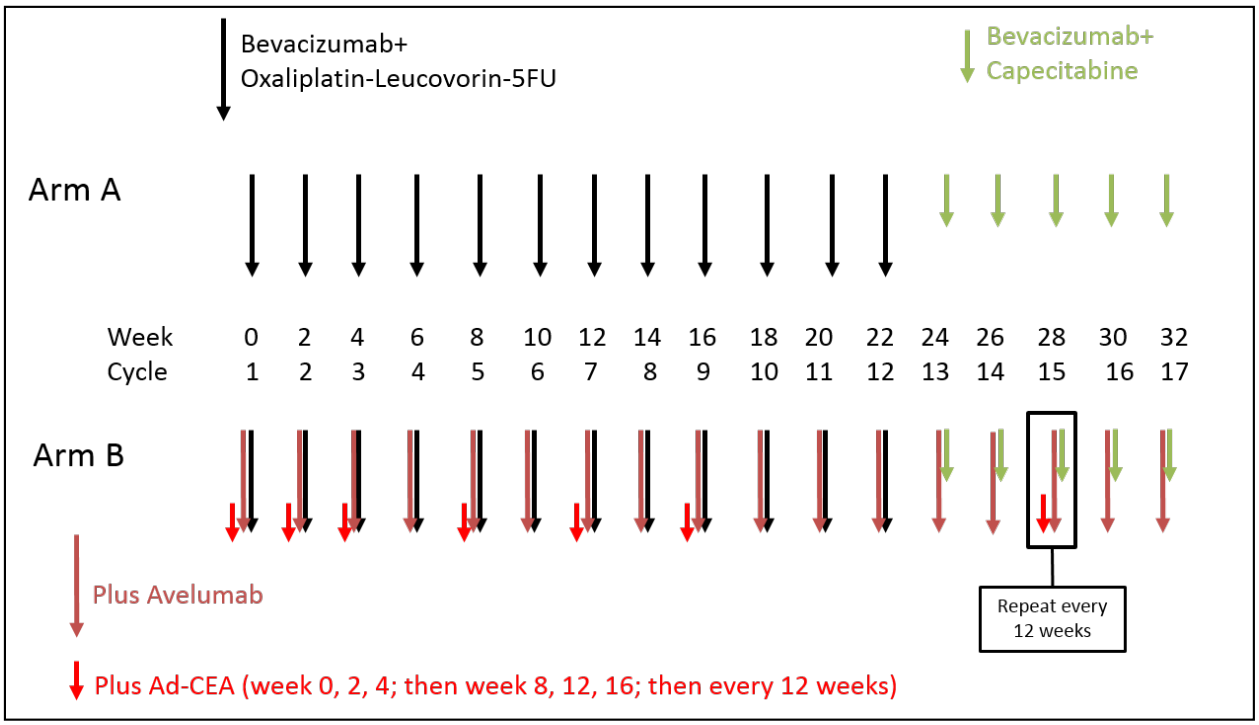
3.1.1.1 Randomization



Note: 5-FU bolus is not given in the FOLFOX-A + Avelumab +Ad-CEA vaccine Arm B

Note: 5-FU may be substituted for capecitabine in the maintenance phase

3.1.1.2 Dosing



Note: 5-FU bolus is not given in the FOLFOX-A + Avelumab +Ad-CEA vaccine Arm B

Note: 5-FU may be substituted for capecitabine in the maintenance phase

3.2 Dose Limiting Toxicity (Lead-In Period Only)

The DLT evaluation period will be defined as the period from the first dose of avelumab until 28 days have passed. DLT will be assessed in the first 6 patients enrolled. Patients will be enrolled at least 5 days from each other to allow adequate assessment of acute toxicity for each patient prior to enrolment of the next patient. If > 1/6 patients experience a DLT, with the combination, the study will be stopped and will not proceed to the randomized phase 2 portion of the study. Subjects who do not complete the DLT observation period for reasons other than a DLT will be replaced.

The goal of the safety evaluation for the combination is to determine if there are any increased or unexpected toxicities due to the combination of therapies that would not be expected with either regimen alone (FOLFOX-A or avelumab). As such, a DLT for the combination will be defined as any adverse event that is unexpected relative to the known safety profile of the standard and investigational agents in the opinion of the investigator. The phase 2.5 portion of the study will not proceed until the DLT evaluation period for 6 patients has been completed.

3.3 Study Stopping Rule (Lead-In Period Only)

Any adverse event meeting criteria for DLT and any death within 30 days of receiving investigational agents, regardless of attribution, will result in the study halting enrollment until an

expedited safety report has been sent to the FDA and the SAE has been evaluated by the investigators.

3.4 DRUG ADMINISTRATION

3.4.1 All Subjects

During the course of the study subjects in study Arm A will receive intravenous (IV) bevacizumab plus FOLFOX (first line therapy) on Days 1 and 2 of a 2 weeks cycle as tolerated per investigator discretion for up to a total of 12 cycles in keeping with standard of care treatment guidelines. Subjects in study Arm B will receive bevacizumab plus FOLFOX (first line therapy) on Days 2 and 3. After completion of first-line induction therapy, both study arm subjects will continue treatment with capecitabine/5-FU plus bevacizumab on a 2-week cycle in the maintenance period. Body surface area (BSA) and dosing will be calculated based on the weight recorded within 2 days prior to avelumab dosing.

3.4.1.1 Administration of Bevacizumab plus FOLFOX (Induction Therapy) [45,46]

Arm A:

- Bevacizumab 5 mg/kg IV over 30 - 90 min on day 1 (infusion rate will be dependent on rate escalation tolerance, section **3.4.1.3**),
- Oxaliplatin 85mg/m^2 IV over 2 hours on day 1 (infusion rate may be adjusted based on tolerance as described in section **3.4.1.4**),
- Leucovorin* 400mg/m^2 IV over 2 hours on day 1,
- 5-FU* 400 mg/m^2 IV bolus on day 1,
- 5-FU* 2400 mg/m^2 IV over 46 hours (+/- 2 hours) to start on day 1.

Arm B:

- Bevacizumab 5 mg/kg IV over 30 - 90 min on day 2 (infusion rate will be dependent on rate escalation tolerance, section **3.4.1.3**),
- Oxaliplatin 85mg/m^2 IV over 2 hours on day 2 (infusion rate may be adjusted based on tolerance as described in section **3.4.1.4**),
- Leucovorin* 400mg/m^2 IV over 2 hours on day 2,
- 5-FU* 2400 mg/m^2 IV over 46 hours (+/- 2 hours) to start on day 2.

* 5-Fluorouracil and leucovorin should be administered separately to avoid the formation of a precipitate. Per package insert, leucovorin is administered first.

* **Note:** Same dose of chemotherapy may be used from previous cycle if patients weight has not changed by more than 10%.

See section **1.2.7.4** for rationale for the removal of 5-FU bolus infusion in Arm B.

3.4.1.2 Administration of Capecitabine plus Bevacizumab (Maintenance Therapy)[43]

- Capecitabine 625 mg/m^2 twice a day every day by mouth. Patients should be instructed to take capecitabine with water within 30 minutes after a meal. Doses should be rounded to the nearest dose achievable without splitting pills.
- Bevacizumab 5 mg/kg IV over 30 - 90 min on day 1, repeat every 14 days (infusion rate will be dependent on rate escalation tolerance, section **3.4.1.3**).
- 5-FU (+/- leucovorin) may be substituted for capecitabine in the maintenance phase. 5FU (+/- leucovorin) dosing during the maintenance phase will be the same as the induction phase (see section **3.4.1.1**).
- **Note:** Same dose of chemotherapy may be used from previous cycle if patient's weight has not changed by more than 10%.

3.4.1.3 Bevacizumab rate escalation

The first dose of bevacizumab will be given over 90 minutes. If the dose is well tolerated with no infusion reactions, the rate will be increased for the second dose, which will be given over 60 minutes. If the second dose is well tolerated, the third dose and all subsequent doses will be given over 30 minutes. If there is an initial infusion reaction at 90 or 60 minute infusion times, but subsequent doses are well-tolerated, the rate may be escalated at the investigator's discretion. Bevacizumab will be permanently discontinued in subjects who experience a grade 3 or 4 infusion reaction suspected to be related to bevacizumab.

3.4.1.4 Oxaliplatin rate

Subjects who experience a grade 1 or 2 hypersensitivity reaction suspected to be related to oxaliplatin, may continue treatment with oxaliplatin at the investigator's discretion. Subsequent infusions must be administered at a slower rate (i.e. over 4 to 6 hours) in addition to premedication (e.g. dexamethasone, ranitidine, and diphenhydramine) 30-60 minutes prior to oxaliplatin.

Subjects who experience a grade 3 or 4 hypersensitivity reaction suspected to be related to oxaliplatin, may continue treatment with oxaliplatin if in the investigator's discretion benefits of continuing therapy outweigh risks. In this situation, the allergy team must be consulted and be in agreement with the plan of care. Skin test and desensitization protocols may be used. However, patients who experience anaphylactic shock or severe immune thrombocytopenia should discontinue oxaliplatin.

3.4.2 Lead-in, Arm A crossovers and Arm B subjects

Subjects in the lead in portion of the study and subjects in Arm B will additionally receive the study drugs, avelumab and Ad-CEA vaccine, during the induction treatment period as well as during the maintenance phase of therapy. Avelumab and Ad-CEA vaccine will be given on day 1 and FOLFOX + bevacizumab will be given on day 2. Subjects in arm A that progress will be offered the opportunity to receive Ad-CEA vaccine and avelumab in combination with a standard chemotherapy regimen as described in section **3.4.2.4**.

3.4.2.1 Avelumab administration

As a routine precaution, subjects enrolled in this trial must be observed for 30 minutes post infusion, in an area with resuscitation equipment and emergency agents. At all times during avelumab treatment, immediate emergency treatment of an infusion-related reaction or a severe hypersensitivity reaction according to institutional standards must be assured. In order to treat possible anaphylactic reactions, dexamethasone 10 mg and epinephrine in a 1:1000 dilution or equivalents should always be available along with equipment for assisted ventilation.

Subjects will receive intravenous infusion of avelumab over 1 hour (-10 minutes / +20 minutes, i.e., 50 to 80 minutes) according to the schedule in sections **3.4.2.3** or **3.4.2.4** as applicable at a dose of 10 mg/kg. Modifications of the infusion rate due to infusion-related reactions are described in section **3.5.1**.

Premedication with an antihistamine and with acetaminophen approximately 30 to 60 minutes prior to the first 4 infusions of avelumab is mandatory (for example, 25-50 mg diphenhydramine and 500-650 mg acetaminophen I.V or oral equivalent). Premedication should be administered for subsequent avelumab infusions based upon clinical judgment and presence/severity of prior infusion reactions. This regimen may be modified based on local treatment standards and guidelines, as appropriate.

The dose of avelumab will be calculated based on the weight of the subject obtained within 2 days prior to avelumab administration. Subjects will receive avelumab at the appropriate schedule until confirmed progression, unacceptable toxicity, or any criterion for withdrawal from the therapy.

Avelumab should be given prior to IV chemotherapy during each cycle, when possible.

Note: Same dose of avelumab may be used from previous cycle if patients weight has not changed by more than 10%.

3.4.2.2 Ad-CEA vaccine administration

Ad-CEA vaccine will be administered as subcutaneous injection in the thigh on Day 1 of specific cycles, prior to avelumab. Ad-CEA will be administered on cycles 1, 2, and 3; 5, 7, and 9; and every 6 cycles thereafter.

Ad-CEA will be provided in a frozen state in a 2 ml vial with a fill volume of 1 ml of extractable vaccine which contains $5x10^{11}$ total virus particles. The product should be stored at –20°C until used and immediately thawed before use.

<u>To administer $5x10^{11}$ virus particles by subcutaneous injection:</u>
Withdraw 1.0 mL of contents from the previously thawed, supplied Ad-CEA from the vial and administer subcutaneously to the patient without any further manipulation.

3.4.2.3 Lead-in and Arm B Induction and Maintenance

During the induction and maintenance phases (FOLFOX plus bevacizumab, and capecitabine plus bevacizumab, respectively), avelumab should be given on day 1 of each cycle. In addition, D1 of each cycle of treatment may be moved +/- 2 days for logistical reasons.

3.4.2.4 Crossovers from Arm A

Subjects that have progressed during the induction phase on Arm A will be given the option to cross over to avelumab + Ad-CEA vaccine in combination FOLFOX in order to exploit the synergistic effects of the combination therapy. Subjects on Arm A who have progressed during the maintenance phase will be given the option to cross over to avelumab + Ad-CEA vaccine in combination with either capecitabine/5-FU + bevacizumab or capecitabine/5-FU+bevacizumab+oxaliplatin based on investigator choice. Subjects that cross over will be treated on a 2 week cycle. Avelumab should be given on day 1 of each cycle and prior to the other agents to avoid giving steroids prior to avelumab. In addition, D1 of each cycle of treatment may be moved +/- 2 days for logistical reasons.

Cross-over will not be offered in the case of rapid or symptomatic progression due to the need for implementation of a standard option with known efficacy.

Definitions:

1. Rapid or symptomatic progression is defined by clinical context including what is commonly termed a "visceral crisis." In these cases, there are increased symptoms of disease progression, laboratory abnormalities due to disease progression, or a combination of both.
2. Radiographic disease progression that is not felt to put a patient in imminent danger by the investigator would meet criteria for standard progression with eligibility for cross-over.

3.5 DOSE DELAYS/MODIFICATIONS

3.5.1 Avelumab

Note: standard of care therapy will be continued during any AE related delay or discontinuation of avelumab.

3.5.1.1 Treatment Modification for Symptoms of avelumab infusion related reactions.

Note: These are just guidelines for consideration and may be deviated from on a case by case basis at the investigator's discretion. When deviating from the below guidelines the investigator should document the reason in the medical record.

NCI-CTCAE Grade	Treatment Modification for Avelumab
Grade 1 – mild Mild transient reaction; infusion interruption not indicated; intervention not indicated.	Decrease the avelumab infusion rate by 50% and monitor closely for any worsening.

NCI-CTCAE Grade	Treatment Modification for Avelumab
Grade 2 – moderate Therapy or infusion interruption indicated but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDs, narcotics, i.v. fluids); prophylactic medications indicated for ≤ 24 hours.	Temporarily discontinue avelumab infusion. Resume infusion at 50% of previous rate once infusion related reaction has resolved or decreased to at least Grade 1 in severity, and monitor closely for any worsening.
Grade 3 or Grade 4 – severe or life-threatening *Grade 3:* Prolonged (e.g., not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence of symptoms following initial improvement; hospitalization indicated for clinical sequelae. *Grade 4*: Life-threatening consequences; urgent intervention indicated.	Stop the avelumab infusion immediately and disconnect infusion tubing from the subject. Subjects have to be withdrawn immediately from avelumab treatment and must not receive any further avelumab treatment.

If avelumab infusion rate has been decreased by 50% or interrupted due to an infusion reaction, it must remain decreased for the next scheduled infusion. If no infusion reaction is observed in the next scheduled infusion, the infusion rate may be returned to baseline at the subsequent infusions based on investigator's medical judgment.

If hypersensitivity reaction occurs, the subject must be treated according to the best available medical practice.

3.5.1.2 Management of Immune-mediated Adverse Reactions

Note: These are just guidelines for consideration and may be deviated from on a case by case basis at the investigator's discretion. When deviating from the below guidelines the investigator should document the reason in the medical record.

Gastrointestinal irAEs		
Severity of Diarrhea/Colitis (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Diarrhea: < 4 stools/day over Baseline Colitis: asymptomatic	Continue avelumab therapy Symptomatic treatment (e.g. loperamide)	Close monitoring for worsening symptoms Educate subject to report worsening immediately If worsens: Treat as Grade 2, 3 or 4.

Grade 2 Diarrhea: 4 to 6 stools per day over Baseline; IV fluids indicated < 24 hours; not interfering with ADL Colitis: abdominal pain; blood in stool	Withhold avelumab therapy Symptomatic treatment	If improves to Grade ≤ 1: Resume avelumab therapy If persists > 5-7 days or recurs: Treat as Grade 3 or 4.
Grade 3 to 4 Diarrhea (Grade 3): ≥ 7 stools per day over Baseline; incontinence; IV fluids ≥ 24 h; interfering with ADL Colitis (Grade 3): severe abdominal pain, medical intervention indicated, peritoneal signs Grade 4: life-threatening, perforation	Withhold avelumab for Grade 3. Permanently discontinue avelumab for Grade 4 or recurrent Grade 3. 1.0 to 2.0 mg/kg/day prednisone IV or equivalent Add prophylactic antibiotics for opportunistic infections Consider lower endoscopy	If improves: Continue steroids until Grade ≤ 1, then taper over at least 1 month; resume avelumab therapy following steroids taper (for initial Grade 3). If worsens, persists > 3 to 5 days, or recurs after improvement: Add infliximab 5mg/kg (if no contraindication). Note: infliximab should not be used in cases of perforation or sepsis.
Dermatological irAEs		
Grade of Rash (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 to 2 Covering ≤ 30% body surface area	Continue avelumab therapy Symptomatic therapy (for example, antihistamines, topical steroids)	If persists > 1 to 2 weeks or recurs: Withhold avelumab therapy Consider skin biopsy Consider 0.5-1.0 mg/kg/day prednisone or equivalent. Once improving, taper steroids over at least 1 month, consider prophylactic antibiotics for opportunistic infections, and resume avelumab therapy following steroids taper. If worsens: Treat as Grade 3 to 4.
Grade 3 to 4 Grade 3: Covering > 30% body surface area; Grade 4: Life threatening consequences	Withhold avelumab for Grade 3. Permanently discontinue for Grade 4 or recurrent Grade 3. Consider skin biopsy Dermatology consult 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections	If improves to Grade ≤ 1: Taper steroids over at least 1 month; resume avelumab therapy following steroids taper (for initial Grade 3).

Pulmonary irAEs		
Grade of Pneumonitis (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Radiographic changes only	Consider withholding avelumab therapy Monitor for symptoms every 2 to 3 days Consider Pulmonary and Infectious Disease consults	Re-assess at least every 3 weeks If worsens: Treat as Grade 2 or Grade 3 to 4.
Grade 2 Mild to moderate new symptoms	Withhold avelumab therapy Pulmonary and Infectious Disease consults Monitor symptoms daily; consider hospitalization 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Consider bronchoscopy, lung biopsy	Re-assess every 1 to 3 days If improves: When symptoms return to Grade ≤ 1, taper steroids over at least 1 month, and then resume avelumab therapy following steroids taper If not improving after 2 weeks or worsening: Treat as Grade 3 to 4.
Grade 3 to 4 Grade 3: Severe new symptoms; New/worsening hypoxia; Grade 4: Life-threatening	Permanently discontinue avelumab therapy. Hospitalize. Pulmonary and Infectious Disease consults. 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Consider bronchoscopy, lung biopsy	If improves to Grade ≤ 1: Taper steroids over at least 1 month If not improving after 48 hours or worsening: Add additional immunosuppression (for example, infliximab, cyclophosphamide, IV immunoglobulin, or mycophenolate mofetil)
Hepatic irAEs		
Grade of Liver Test Elevation (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Grade 1 AST or ALT > ULN to 3.0 x ULN and/or Total bilirubin > ULN to 1.5 x ULN	Continue avelumab therapy	Continue liver function monitoring If worsens: Treat as Grade 2 or 3 to 4.
Grade 2 AST or ALT > 3.0 to ≤ 5 x ULN and/or total bilirubin > 1.5 to ≤ 3 x ULN	Withhold avelumab therapy Increase frequency of monitoring to every 3 days.	If returns to Grade ≤ 1: Resume routine monitoring; resume avelumab therapy. If elevation persists > 5 to 7 days or worsens: Treat as Grade 3 to 4.

Grade 3 to 4 AST or ALT > 5 x ULN and/or total bilirubin > 3 x ULN	Permanently discontinue avelumab therapy Increase frequency of monitoring to every 1 to 2 days 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Consult gastroenterologist/ hepatologist Consider obtaining MRI/CT scan of liver and liver biopsy if clinically warranted	If returns to Grade ≤ 1: Taper steroids over at least 1 month If does not improve in > 3 to 5 days, worsens or rebounds: Add mycophenolate mofetil 1 gram (g) twice daily If no response within an additional 3 to 5 days, consider other immunosuppressants per local guidelines.
Renal irAEs		
Grade of Creatinine Increased (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Creatinine increased > ULN to 1.5 x ULN	Continue avelumab therapy	Continue renal function monitoring If worsens: Treat as Grade 2 to 3 or 4.
Grade 2 to 3 Creatinine increased > 1.5 and ≤ 6 x ULN	Withhold avelumab therapy Increase frequency of monitoring to every 3 days 1.0 to 2.0 mg/kg/day prednisone or equivalent. Add prophylactic antibiotics for opportunistic infections Consider renal biopsy	If returns to Grade ≤1: Taper steroids over at least 1 month, and resume avelumab therapy following steroids taper. If worsens: Treat as Grade 4.
Grade 4 Creatinine increased > 6 x ULN	Permanently discontinue avelumab therapy Monitor creatinine daily 1.0 to 2.0 mg/kg/day prednisone or equivalent. Add prophylactic antibiotics for opportunistic infections Consider renal biopsy Nephrology consult	If returns to Grade ≤1: Taper steroids over at least 1 month.

Cardiac irAEs		
Myocarditis	**Initial Management**	**Follow-up Management**
New onset of cardiac signs or symptoms and / or new laboratory cardiac biomarker elevations (e.g. troponin, CK-MB, BNP) or cardiac imaging abnormalities suggestive of myocarditis.	Withhold avelumab therapy. Hospitalize. In the presence of life threatening cardiac decompensation, consider transfer to a facility experienced in advanced heart failure and arrhythmia management. Cardiology consult to establish etiology and rule-out immune-mediated myocarditis. Guideline based supportive treatment as per cardiology consult.* Consider myocardial biopsy if recommended per cardiology consult.	If symptoms improve and immune-mediated etiology is ruled out, re-start avelumab therapy. If symptoms do not improve/worsen, viral myocarditis is excluded, and immune-mediated etiology is suspected or confirmed following cardiology consult, manage as immune-mediated myocarditis.
Immune-mediated myocarditis	Permanently discontinue avelumab. Guideline based supportive treatment as appropriate as per cardiology consult.* 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections.	Once improving, taper steroids over at least 1 month. If no improvement or worsening, consider additional immunosuppressants (e.g. azathioprine, cyclosporine A).
*Local guidelines, or e.g. ESC or AHA guidelines ESC guidelines website: https://www.escardio.org/Guidelines/Clinical-Practice-Guidelines AHA guidelines website: http://professional.heart.org/professional/GuidelinesStatements/searchresults.jsp?q=&y=&t=1001		
Endocrine irAEs		
Endocrine Disorder	**Initial Management**	**Follow-up Management**
Grade 1 or Grade 2 endocrinopathies (hypothyroidism, hyperthyroidism, adrenal insufficiency, type I diabetes mellitus)	Continue avelumab therapy Endocrinology consult if needed Start thyroid hormone replacement therapy (for hypothyroidism), anti-thyroid treatment (for hyperthyroidism), corticosteroids (for adrenal insufficiency) or insulin (for Type I diabetes mellitus) as appropriate. Rule-out secondary endocrinopathies (i.e. hypopituitarism/ hypophysitis)	Continue hormone replacement/suppression and monitoring of endocrine function as appropriate.

Grade 3 or Grade 4 endocrinopathies (hypothyroidism, hyperthyroidism, adrenal insufficiency, type I diabetes mellitus)	Withhold avelumab therapy Consider hospitalization Endocrinology consult Start thyroid hormone replacement therapy (for hypothyroidism), anti-thyroid treatment (for hyperthyroidism), corticosteroids (for adrenal insufficiency) or insulin (for type I diabetes mellitus) as appropriate. Rule-out secondary endocrinopathies (i.e. hypopituitarism/ hypophysitis)	Resume avelumab once symptoms and/or laboratory tests improve to Grade ≤ 1 (with or without hormone replacement/suppression). Continue hormone replacement/suppression and monitoring of endocrine function as appropriate.
Hypopituitarism/Hypophysitis (secondary endocrinopathies)	If secondary thyroid and/or adrenal insufficiency is confirmed (i.e. subnormal serum FT4 with inappropriately low TSH and/or low serum cortisol with inappropriately low ACTH): • Refer to endocrinologist for dynamic testing as indicated and measurement of other hormones (FSH, LH, GH/IGF-1, PRL, testosterone in men, estrogens in women) • Hormone replacement/suppressive therapy as appropriate • Perform pituitary MRI and visual field examination as indicated **If hypophysitis confirmed:** • Continue avelumab if mild symptoms with normal MRI. Repeat the MRI in 1 month • Withhold avelumab if moderate, severe or life-threatening symptoms of hypophysitis and/or abnormal MRI. Consider hospitalization. Initiate corticosteroids (1 to 2 mg/kg/day prednisone or equivalent) followed by corticosteroids taper during at least 1 month. • Add prophylactic antibiotics for opportunistic infections.	Resume avelumab once symptoms and hormone tests improve to Grade ≤ 1 (with or without hormone replacement). In addition, for hypophysitis with abnormal MRI, resume avelumab only once shrinkage of the pituitary gland on MRI/CT scan is documented. Continue hormone replacement/suppression therapy as appropriate.
Other irAEs (not described above)		

Grade of other irAEs (NCI-CTCAE v5)	Initial Management	Follow-up Management
Grade 2 or Grade 3 clinical signs or symptoms suggestive of a potential irAE	Withhold avelumab therapy pending clinical investigation	If irAE is ruled out, manage as appropriate according to the diagnosis and consider re-starting avelumab therapy If irAE is confirmed, treat as Grade 2 or 3 irAE.
Grade 2 irAE or first occurrence of Grade 3 irAE	Withhold avelumab therapy 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Specialty consult as appropriate	If improves to Grade ≤ 1: Taper steroids over at least 1 month and resume avelumab therapy following steroids taper.
Recurrence of same Grade 3 irAEs	Permanently discontinue avelumab therapy 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Specialty consult as appropriate	If improves to Grade ≤ 1: Taper steroids over at least 1 month.
Grade 4	Permanently discontinue avelumab therapy 1.0 to 2.0 mg/kg/day prednisone or equivalent and/or other immunosuppressant as needed Add prophylactic antibiotics for opportunistic infections Specialty consult.	If improves to Grade ≤ 1: Taper steroids over at least 1 month
Requirement for 10 mg per day or greater prednisone or equivalent for more than 12 weeks for reasons other than hormonal replacement for adrenal insufficiency **Persistent Grade 2 or 3 irAE lasting 12 weeks or longer**	Permanently discontinue avelumab therapy Specialty consult	

Abbreviations: ACTH=adrenocorticotropic hormone; ADL=activities of daily living; BNP=B-type natriuretic peptide; CK-MB=creatine kinase MB; FSH=follicle-stimulating hormone; GH=growth hormone; IGF-1=insulin-like growth factor 1; LH=luteinizing hormone; PRL=prolactin.

3.5.2 Active Immunotherapy with Ad-CEA vaccine will be discontinued for:

3.5.2.1 Life-threatening anaphylactic reactions related to the Ad-CEA vaccine.

3.5.2.2 DLT related to active immunotherapy with Ad-CEA as follows: If one or more patients develop a Grade 4 allergic reaction without a clear attributable cause, other than study vaccine.

3.5.3 Bevacizumab

Note: These are just guidelines for consideration and may be deviated from on a case by case basis at the investigator's discretion. When deviating from the below guidelines the investigator should document the reason in the medical record.

3.5.3.1 Hypertension

3.5.3.1.1 Bevacizumab should not be administered unless SBP< 150 and DBP < 90. Management of hypertension is outlined below.

3.5.3.1.2 Mild/Moderate Hypertension

For SBP > 140 and < 210 mm Hg or DBP > 90 and < 120 mm Hg that is sustained over at least a two week period, initiate or adjust anti-hypertensive therapy. Bevacizumab should be delayed until the blood SBP< 150 and DBP < 90.

3.5.3.1.3 Severe Hypertension

For SBP ≥ 210 mm Hg or DBP ≥ 120 mm Hg but without end organ damage, begin anti-hypertensive therapy. Bevacizumab should be delayed until blood SBP< 150 and DBP < 90.

3.5.3.1.4 Hypertensive Urgency or Emergency

Bevacizumab therapy should be permanently discontinued in the presence of hypertensive urgency (DBP >120 with evidence of optic disc edema or progressive end-organ complications) or hypertensive emergency (SBP > 210 and DBP > 120 presenting with headaches, blurred vision, or focal neurological symptoms, or papilledema). .

3.5.3.2 Thrombotic Events

3.5.3.2.1 Deep Vein Thrombosis

Bevacizumab should not be administered in the presence of deep vein thrombosis or pulmonary embolism. Anticoagulation may be used and bevacizumab may be restarted when thrombosis is resolved.

3.5.3.2.2 Arterial Thromboembolic Events

Bevacizumab should be permanently discontinued in the presence of new arterial thrombotic disease, including but not limited to, myocardial infarct due to coronary thrombosis and ischemic stroke.

3.5.3.3 Neutropenia

Bevacizumab should be held if the absolute neutrophil count (ANC) is less than 750 cells/μL until ANC is again above 750 cells/μL.

3.5.3.4 Hemorrhage

3.5.3.4.1 Life Threatening

In the case of hemoptysis, hematemesis, hematochezia, intracranial hemorrhage or any significant blood loss, bevacizumab should not be administered.

3.5.3.5 Proteinuria

If urine dipstick indicates ≥2+ proteinuria, bevacizumab administration should be delayed pending the results of a 24-hour urine collection for protein. If the 24-hour urine protein is ≥2 grams, bevacizumab will be delayed for up to 6 weeks, until the proteinuria is <2+ or the 24-hour urine protein is < 1 grams/24 hr.

3.5.3.6 Liver Function Abnormalities

Bevacizumab should be withheld in the event of ≥ Grade 3 LFT elevations attributed to bevacizumab and should not resume until the abnormalities have recovered to ≤ Grade 1.

3.5.3.7 Allergic Reaction

In case of flushing, shortness of breath, facial edema, headache, chills, back pain, tightness of the chest and throat, and/or hypotension, fever or rash, the infusion should be suspended until the patient is assessed until the events have subsided. For grade 3 or 4 allergic reactions, bevacizumab should be permanently discontinued.

3.5.3.8 Surgical or periodontal procedures

If there is need for an elective major surgical or periodontal procedure, bevacizumab should be held beginning at least 6 weeks prior to the procedure and must not be resumed before 4 weeks after the surgical procedure. For urgent or emergent surgery or endoscopic procedures, bevacizumab should be held for 4 weeks after the procedure. Longer delays may be necessary, if clinically indicated, in order to ensure that adequate healing has taken place prior to bevacizumab resumption. For minor procedures (i.e. interventional radiology diagnostic procedures), bevacizumab will be held only if there is evidence that healing is compromised.

3.5.3.9 Reversible Posterior Leukoencephalopathy Syndrome (RPLS)

Bevacizumab should not be administered to patients with signs or symptoms of RPLS. Evaluation should include neurologic evaluation, ocular examination, head MRI, and blood pressure assessment. If these clinical criteria are consistent with the diagnosis of RPLS, bevacizumab will be permanently discontinued.

3.5.3.10 Gastrointestinal Perforations and Fistula Formation

Bevacizumab should be discontinued in the event of a gastrointestinal perforation or the formation of a fistula.

3.5.3.11 Miscellaneous

Bevacizumab may be held at any point when in the opinion of the investigator the risks of giving bevacizumab outweigh the potential benefit.

3.5.4 Capecitabine

Dose adjustments for toxicities per CTCAE as per table.

Note: These are just guidelines for consideration and may be deviated from on a case by case basis at the investigator's discretion. When deviating from the below guidelines the investigator should document the reason in the medical record.

Summary of management of common Capecitabine toxicities	
Toxicity	**Capecitabine dose modification**
Grade 2 hand foot syndrome	Interrupt until ≤ grade 1. May then restart capecitabine at full dose. For second occurrence, hold capecitabine until ≤ grade 1, then restart capecitabine one dose level lower (75% of starting dose first occurrence, 50% of the starting dose the second occurrence, discontinue the third occurrence).
Grade 3 hand foot syndrome	Interrupt until ≤ grade 1. Then restart capecitabine one dose level lower (75% of starting dose first occurrence, 50% of the starting dose the second occurrence, discontinue the third occurrence).
Grade 2 Nausea/Vomiting	Continue at current dose, modify anti-emetic regimen
Grade 3 Nausea/Vomiting	Hold until recovery to grade 1 (or grade 2 if present at baseline) then resume one dose level lower*
Grade 2 Diarrhea	Continue current dose, add optimal anti-diarrheal therapy
Grade 3 (or greater) Diarrhea	Hold until recovery to grade 1 (or grade 2 if present at baseline) then resume one dose level lower*
Grade 3 (or greater) fatigue	Hold until recovery to grade 1 (or grade 2 if present at baseline) then resume one dose level lower*
Grade 3 (or greater) mucositis	Hold until recovery to grade 1 (or grade 2 if present at baseline) then resume one dose level lower*
Grade 2 thrombocytopenia	Continue at current dose
Grade 3 thrombocytopenia	Hold until recovery until ≤ grade 1 then resume one dose level lower*

Summary of management of common Capecitabine toxicities	
Toxicity	**Capecitabine dose modification**
Grade 4 thrombocytopenia	Hold until recovery until ≤ grade 1 then resume one dose level lower*
Grade 3 neutropenia	Hold until recovery until ≤ grade 1 then resume one dose level lower*
Grade 4 neutropenia	Hold until recovery until ≤ grade 1 then resume one dose level lower*
≥ Grade 3 febrile neutropenia	Hold until resolution of fever and neutropenia to ≤ grade 1. Then resume on dose level lower*
*Dose reductions for capecitabine 1. Starting dose: 625 mg/m^2 BID continuously 2. DL -1: 470 mg/m^2 BID continuously 3. DL -2: 310 mg/m^2 BID continuously 4. Discontinuation of capecitabine.	
Capecitabine may be held in the maintenance setting (i.e., chemo holiday) when in the opinion of the investigator the risks of giving capecitabine outweigh the potential benefit. The same is true for 5-FU when given as an alternative in the maintenance setting.	

3.5.5 FOLFOX dose adjustment

Note: These are just guidelines for consideration and may be deviated from on a case by case basis at the investigator's discretion. When deviating from the below guidelines the investigator should document the reason in the medical record.

For Arm A

	Adverse Event	First Occurrence	Second Occurrence	Third Occurrence	Fourth Occurrence
Hematologic Toxicity	ANC <1000/mL OR Platelets <75000/mL OR Any Febrile Neutropenia	1. Hold all therapy and check CBC weekly until recovery to: ANC ≥1000/mL *and* Platelets ≥75000/mL AND 2. Discontinue 5-FU bolus on Day 1 at the next cycle and consider adding growth factor support for neutropenia	1. Hold all therapy and check CBC weekly until recovery to: ANC ≥1000/mL *and* Platelets ≥75000/mL AND 2. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	1. Hold all therapy and check CBC weekly until recovery to: ANC ≥1000/mL *and* Platelets ≥75000/mL AND 2. Decrease 5-FU continuous infusion to 1800mg/m2 *and* decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	1. Hold all therapy and check CBC weekly until recovery to: ANC ≥1000/mL *and* Platelets ≥75000/mL AND 2. Decrease 5-FU continuous infusion to 1200mg/m2 *and* discontinue Oxaliplatin on Day 1 at the next cycle
	ANC ≥1000/ml but <1500/mL OR Platelets ≥75000/ml but <100000/mL	1. Discontinue 5-FU bolus on Day 1 at the next cycle and consider adding growth factor support for neutropenia	1. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	1. Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	1. Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle
	Any other CTC ≥ Grade 3 hematologic toxicity, (e.g. HgB <8.0g/dL)	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Discontinue 5-FU bolus on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle
Non-Hematologic Toxicity	Nausea/Vomiting CTC ≥ Grade 3	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Add a aprepitant if not already receiving. If already receiving, consider adding olanzapine and/or administering dexamethasone 4mg PO 24 and 48 hrs after oxaliplatin	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Discontinue 5-FU bolus and reduce oxaliplatin to 65mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1800mg/m2 and decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1200mg/m2 *and* discontinue Oxaliplatin on Day 1 at the next cycle
	Diarrhea CTC ≥ Grade 3, despite optimal antidiarrheal therapy	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Discontinue 5-FU bolus at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1200mg/m2 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle

	Fatigue CTC ≥ Grade 3 OR Mucositis CTC ≥ Grade 3 OR Other Toxicity Not Defined Herein CTC ≥ Grade 3 despite optimal supportive care	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Discontinue 5-FU bolus on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1200mg/m2 and decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle
FOLFOX-Specific Toxicities	Peripheral Neuropathy CTC ≥ Grade 2	1. Hold all therapy until recovery to Grade ≤ 2 AND 2. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade ≤ 2 AND 2. Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade ≤ 2 AND 2. Discontinue Oxaliplatin on Day 1 at the next cycle	**N/A**
	Hand-Foot syndrome CTC ≥ Grade 3	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Discontinue 5-FU bolus on Day 1 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease 5-FU continuous infusion to 1200mg/m2 at the next cycle	1. Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND 2. Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle

For Arm B

	Adverse Event	First Occurrence	Second Occurrence	Third Occurrence	Fourth Occurrence
Hematologic Toxicity	ANC <1000/mL OR Platelets <75000/mL OR Any Febrile Neutropenia	Hold all therapy and check CBC weekly until covery to: ANC ≥1000/mL *and* Platelets ≥75000/mL 3. For thrombocytopenia: Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle For neutropenia: Add growth factor support	Hold all therapy and check CBC weekly until covery to: ANC ≥1000/mL *and* Platelets ≥75000/mL For thrombocytopenia: Decrease Oxaliplatin to 5mg/m2 on Day 1 at the next cycle For neutropenia: Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy and check CBC weekly until recovery to: ANC ≥1000/mL *and* Platelets ≥75000/mL For thrombocytopenia: Decrease 5-FU continuous infusion to 1800mg/m2 For neutropenia: Decrease 5-FU continuous infusion to 1800mg/m2 *and* decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	Hold all therapy and check CBC weekly until recovery to: ANC ≥1000/mL *and* Platelets ≥75000/mL Decrease 5-FU continuous infusion to 1200mg/m2 *and* discontinue Oxaliplatin on Day 1 at the next cycle
	ANC ≥1000/ml but <1500/mL OR Platelets ≥75000/ml but <100000/mL	For thrombocytopenia: Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle For neutropenia: Add growth factor support	For thrombocytopenia: Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle For neutropenia: Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	For thrombocytopenia: Decrease 5-FU continuous infusion to 1800mg/m2 For neutropenia: Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	For thrombocytopenia: Decrease 5-FU continuous infusion to 1200mg/m2 For neutropenia: Decrease 5-FU continuous infusion to 1800mg/m2
	Any other CTC ≥ Grade 3 hematologic toxicity, (e.g. HgB <8.0g/dL)	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1800 mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1200mg/m2 on Day 1 at the next cycle

Non-Hematologic Toxicity	Nausea/Vomiting CTC ≥ Grade 3	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Add a aprepitant if not already receiving. If already receiving, consider adding olanzapine and/or administering dexamethasone 4mg PO days at 24 and 48 hrs after oxaliplatin.	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Reduce oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1800mg/m2 and decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1200mg/m2 *and* discontinue Oxaliplatin on Day 1 at the next cycle
	Diarrhea CTC ≥ Grade 3, despite optimal antidiarrheal therapy	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1200mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle
	Fatigue CTC ≥ Grade 3 OR Mucositis CTC ≥ Grade 3 OR Other Toxicity Not Defined Herein CTC ≥ Grade 3 despite optimal supportive care	Hold all therapy until recovery to Grade 1 (or rade 2 level, if present at baseline) AND Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1200mg/m2 on Day 1 at the next cycle
FOLFOX-Specific Toxicities	Peripheral Neuropathy CTC ≥ Grade 2	Hold all therapy until recovery to Grade ≤ 2 AND Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade ≤ 2 AND Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade ≤ 2 AND Discontinue Oxaliplatin on Day 1 at the next cycle	N/A
	Hand-Foot syndrome CTC ≥ Grade 3	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1800mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease 5-FU continuous infusion to 1200mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 65mg/m2 on Day 1 at the next cycle	Hold all therapy until recovery to Grade 1 (or Grade 2 level, if present at baseline) AND Decrease Oxaliplatin to 50mg/m2 on Day 1 at the next cycle

3.6 STUDY CALENDAR

- Weight may be assessed within 2 days prior to avelumab dosing for all cycles
- Cycle 1 day 1 assessments may be performed within 16 days prior to what is indicated
- Other day 1 assessments in cycles 2 and beyond may be performed within 1 week prior to indicated time
- **D1 and D2 of each cycle may be moved +/- 2 days for logistical reasons**
- **When a cycle must be delayed for logistical or medical reasons that cycle can be given the following week (as opposed to waiting 2 weeks) and future dosing should reset to every other week following that administration.**

			Induction Therapy												Maintenance Therapy	Post Therapy Follow-up	
Procedure	**Screen-ing[1]**	**Base-line[2]**	**C1 D1[3]**	**C2 D1**	**C3 D1**	**C4 D1**	**C5 D1**	**C6 D1**	**C7 D1**	**C8 D1**	**C9 D1**	**C10 D1**	**C11 D1**	**C12 D1**	**CX D1**	**30 day safety visit[4]**	**Long term follow up**
Bevacizumab[12]			X	X	X	X	X	X	X	X	X	X	X	X	X		
FOLFOX[13]			X	X	X	X	X	X	X	X	X	X	X	X			
Capecitabine/5-FU															X		
Avelumab [14]			X	X	X	X	X	X	X	X	X	X	X	X	X		
Ad-CEA vaccine [14]			X	X	X		X		X		X				X		
History and PE	X		X	X	X	X	X	X	X	X	X	X	X	X	Every 4 cycles	X	
Height		X															
Weight		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Vital signs	X														X	X	
Performance Score	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	
CBC with differential	X	X	X[2]	X	X	X	X	X	X	X	X	X	X	X	X	X	
Hepatic +Acute Care Panels[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Mineral Panel[4]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
ACTH, ANA, RF, TSH, T4[6]		X	X				X				X				Every 4 cycles	X	
PT/INR, PTT	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Urinalysis[7]	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	

Procedure	Screen-ing[1]	Base-line[2]	Induction Therapy												Maintenance Therapy	Post Therapy Follow-up	
			C1 D1[3]	C2 D1	C3 D1	C4 D1	C5 D1	C6 D1	C7 D1	C8 D1	C9 D1	C10 D1	C11 D1	C12 D1	CX D1	30 day safety visit[4]	Long term follow up
12 lead EKG	X		X		X		X				X				Every 4 cycles		
HBV, HCV and HIV testing	X																
Serum or urine HCG in women of childbearing potential	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Pathology confirmation of dx	X																
HLA profiling, HLA A, B, C subtype		X															
Archival tumor tissue	X																
Optional Biopsy[8]		X					X	→	→	→	→	→	→	→	→		
Correlative research blood[9]			X		X		X				X				Every 4 cycles (with restaging)		
CT CAP or CT chest + MRI abdomen/pelvis	X						X				X				Every 4 cycles		
Brain MRI if clinically indicated	X																
Adverse Events			X	→	→	→	→	→	→	→	→	→	→	→	→		
Concomitant Medications			X	→	→	→	→	→	→	→	→	→	→	→	→		
Consent	X																
NIH Advanced Directives Form[10]		X															

Procedure	Screen-ing[1]	Base-line[2]	Induction Therapy												Maintenance Therapy	Post Therapy Follow-up	
			C1 D1[3]	C2 D1	C3 D1	C4 D1	C5 D1	C6 D1	C7 D1	C8 D1	C9 D1	C10 D1	C11 D1	C12 D1	CX D1	30 day safety visit[4]	Long term follow up
Annual phone call for survival[11]																	X

[1] Screening assessments are performed within 16 days prior to enrollment **except** for: scans - done within 28 days prior to enrollment; viral screens (HIV, HBV, HCV) – done within 3 months prior to enrollment; confirmation of dx which requires specimens collected within 6 month prior to study enrollment

[2] May be performed within 16 days prior to initiation of study therapy.

[3] Cycle 1 day 1 assessments may be performed within 16 days prior to what is indicated.

[4] 30 day safety visit occurs approximately 4 – 5 weeks after the last administration of study drug regardless of reason for discontinuation. Information may be obtained by telephone if patient refuses visit.

[5] **Acute Care Panel:** Sodium (NA), Potassium (K), Chloride (CL) Total CO2 (Bicarbonate), Creatinine, Glucose, Urea nitrogen, eGFR; **Hepatic Panel:** Alkaline Phosphatase, ALT/GPT, AST/GOT, Total Bilirubin, Direct Bilirubin; **Mineral Panel:** Albumin, Calcium, Magnesium (Mg), Phosphorus

[6] **ACTH**: adrenocorticotropic hormone; **ANA:** anti-nuclear antibody; **RF:** rheumatoid factor; **TSH:** Thyroid-stimulating hormone; **T4:** Free thyroxine.

[7] Only urine dipstick required initially. If positive for protein content, a 24 hour urine collection for protein will be performed.

[8] Two optional biopsies can be performed: one at Baseline and second at any time after completion of C4 per PI discretion.

[9] Correlative blood samples: baseline sample: 2.5 mL in PAX gene tube; On therapy: blood collected pre-dose: 5 (10mL) green top sodium heparin tubes for PBMC; 1 (8mL) SST tube for serum samples. Samples will be collected in selected patients at the investigator's discretion.

[10] As indicated in section **12.3**, all subjects will be offered the opportunity to complete an NIH advanced directives form. This should be done preferably at baseline but can be done at any time during the study as long as the capacity to do so is retained. The completion of the form is strongly recommended, but is not required.

[11] Patients who have come off treatment for disease progression will be followed by phone annually for survival. Patients who have not progressed on treatment will continue to be followed and scanned per investigator discretion until progression.

[12] On Day 1 in Arm A and Day 2 in Arm B (Day 1 for Arm B maintenance)

[13] On Days 1 and 2 in Arm A and Days 2 and 3 in Arm B

[14] Only Arm B; While avelumab will be given at every cycle, Ad-CEA will only be given on C 1, 2, 3, 5, 7, 9 and every 6 cycles (approximately 12 weeks) thereafter.

3.7 CRITERIA FOR REMOVAL FROM PROTOCOL THERAPY AND OFF STUDY CRITERIA

Prior to removal from study, effort must be made to have all subjects complete a safety visit approximately 30 days following the last dose of study therapy.

3.7.1 Criteria for removal from protocol therapy

- Completion of protocol therapy.
- Progressive disease requiring a change in systemic therapy (exception: patients Arm A that have opted to cross over to Avelumab + Ad-CEA vaccine + standard of care).
- Participant requests to be withdrawn from active therapy.
- Unacceptable Toxicity as defined as any serious adverse event that is unexpected relative to the known safety profile of the investigational agents in the opinion of the investigator, many of which are described in sections **3.5** and **3.2.**
- Start of another systemic anticancer treatment or participation in another investigational therapeutic trial. Focal palliative radiotherapy, ablation, or surgery to a site of disease will not necessitate removal from protocol therapy. Although where appropriate progressive disease per RECIST v1.1 should be clearly documented and the decision to continue treatment beyond PD should be noted in the medical records (e.g. overall response to treatment except for isolated progressive lesion which has been treated by local intervention).
- Positive pregnancy test.
- Investigator discretion.

Regardless of reason for removal from study therapy, patients will be asked to have a 30 day follow up safety visit. Patients who refuse to return for this visit will be asked to review any safety concerns by phone within this time period.

3.7.2 Off-Study Criteria

- Screen Failure
- Investigator discretion
- Participant requests to be withdrawn from study
- Death
- PI decision to end the study

3.7.3 Clinical Center Site Off Protocol Therapy and Off-Study Procedure

Authorized staff must notify Central Registration Office (CRO) when a subject is taken off protocol therapy and when a subject is taken off-study. A Participant Status Update Form from the web site (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) main page must be completed and sent via encrypted email to: NCI Central Registration Office ncicentralregistration-l@mail.nih.gov .

3.7.4 Participating Site Off Therapy and Off-Study Procedure

4 THE PARTICIPANT STATUS UPDATE FORM WILL BE SUPPLIED BY THE CCR STUDY COORDINATOR. SEND THE COMPLETED FORM TO THE CCR STUDY COORDINATOR.CONCOMITANT MEDICATIONS/MEASURES

4.1 SUPPORTIVE CARE

4.1.1 Recommended Management for Severe Hypersensitivity Reaction and Flu-like Symptoms

Management

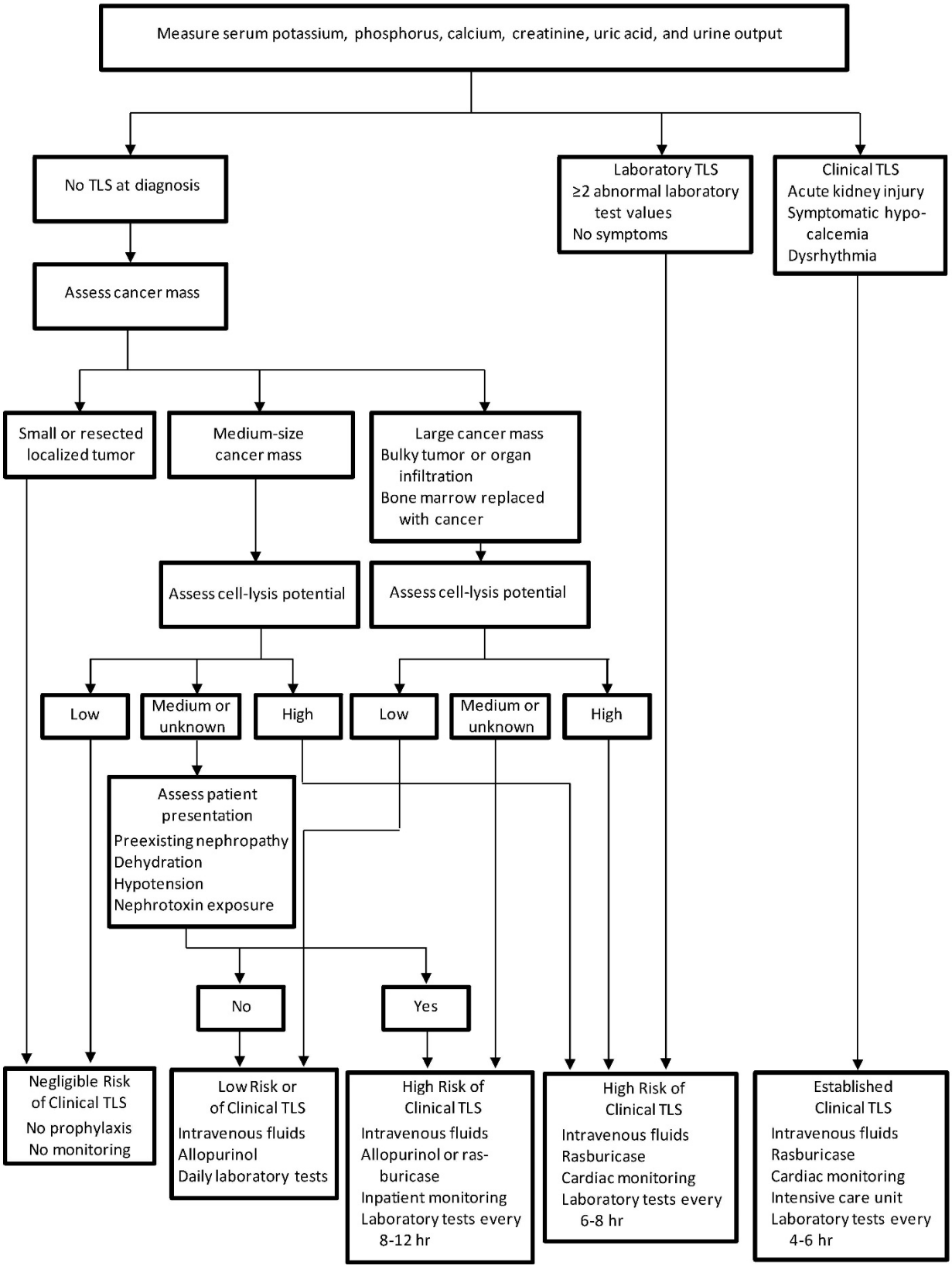
1. Epinephrine injection and dexamethasone infusion (or equivalent)
2. Patient should be placed on monitor immediately
3. Alert intensive care unit (ICU) for possible transfer if required

For prophylaxis of flu-like symptoms, 25 mg indomethacin or comparable NSAID dose (e.g., ibuprofen 600 mg, naproxen sodium 500 mg) may be administered 2 hours before and 8 hours after the start of each dose of avelumab i.v. infusion. Alternative treatments for fever (e.g., acetaminophen) may be given to subjects at the discretion of the investigator.

4.1.2 Recommendations for the management of tumor lysis syndrome

Since avelumab can induce ADCC, there is a potential risk of tumor lysis syndrome. Should this occur, subjects should be treated as per local guidelines and the management algorithm (**Figure 23**) published by Howard et al.[74]

Figure 23. Assessment and Initial Management of Tumor Lysis Syndrome (TLS)



4.1.3 General supportive care

Additional supportive care guidelines for avelumab are found in the management tables in section **3.5.1.2**

Because there is a potential for interaction of capecitabine with other concomitantly administered drugs through the CYP2C9 system, the case report form (CRF) must capture the concurrent use of all other drugs, over-the-counter medications, or alternative therapies. The Principal Investigator should be alerted if the patient is taking any agent known to affect or with the potential to interact with capecitabine. Patients will be transitioned to an acceptable alternative if available. Common examples include aluminum and magnesium hydroxide containing antacids, coumadin, and phenytoin.

Diarrhea will be managed with the following regimen: loperamide (4 mg PO) at onset of symptoms, followed by 2 mg loperamide every 2 hours while awake (or 4 mg PO every 4 hours while sleeping) up to a maximum of 16 mg loperamide per day. Additional agents may be used concurrently if loperamide is not adequate to control diarrhea as a single agent.

Nausea and vomiting will be managed through the use of appropriate simple supportive measures. Recommended premedication for oxalaplatin includes aprepitant 125mg PO, dexamethasone 12mg PO, ondansetron 24mg PO and aprepitant 80mg 24 and 48 hours after receiving oxaliplatin. Alternative options may be considered, including 10-20mg IV dexamethasone and 16mg ondansetron intravenously 30-60 minutes prior to infusion, for example.

Patients will receive full supportive care per NIH CC guidelines as needed while on this study including blood product support, intravenous hydration, antibiotic treatment and treatment of other newly diagnosed or concurrent medical conditions.

Patients should be advised to drink plenty of water or take rehydration fluids to avoid dehydration if diarrhea occurs.

5 BIOSPECIMEN COLLECTION

5.1 CORRELATIVE STUDIES FOR RESEARCH/PHARMACOKINETIC STUDIES

5.1.1 IMMUNOLOGICAL STUDIES

Samples from patients enrolled at Georgetown and the Clinical Center will be available for immunologic correlative studies as described below. Samples will be collected in selected patients at the investigator's discretion.

5.1.1.1 Peripheral blood

Five (10mL) green top sodium heparin tubes for PBMC; 1 (8mL) SST tubes for serum samples will be drawn on day 1 of Cycle 1, 3, 5, 9 and every 4 cycles thereafter as described in the **Study Calendar**. The following assessments are planned in the Laboratory of Tumor Immunology and Biology (LTIB), CCR, NCI for the samples collected. Please see section **5.1.1.3.1** for sample processing information.

5.1.1.1.1 Leukocyte subpopulations and immune activation status:

Leukocyte subpopulations and immune activation status will be assessed by flow cytometry (FACS) on PBMC. Multiparameter FACS analysis using combination of markers will be performed to characterize leukocyte subpopulations and their functional state such as:

- T-cells subsets and activation state
- T-cells functionality
- PD-1 signaling pathway
- Regulatory T-cells
- Other leukocytes subsets including B-cells, monocytes
- NK cells and related cytotoxicity
- Differentiation stage of T-cells subsets
- Myeloid derived suppressor cells and APCs

5.1.1.1.2 Comparison presence of TAA-specific T lymphocytes using TAA-specific peptides will be performed on pre- and post-treatment samples if enough PBMCs are available

5.1.1.1.3 Further exploratory markers related to the mechanism of action of the drug such as soluble PD-L1 sera level, cytokine profile, and auto-antigen proteomic arrays may be explored.

5.1.1.1.4 T Cell Clonal Expansion Assay

cDNA from PBMC will be amplified using locus specific primers for TCR-β. Previously described methods will be used to map the V region and identify the J region (Klinger) and identify clonal sequences of interest. Analytics tools will be used to sort and identify clonal populations of interest in the post- versus pre-treatment samples. Correlation of expansion of a clonal TCR population post treatment will be correlated with clinical outcomes.

5.1.1.1.5 Serum soluble factors and ADCC-related side effects (such as in vitro ADCC activity assays, sCD40L and/or sCD27) may be explored.

5.1.1.2 Tissue analysis (Optional)

Patents will be asked to provide archival tumor sample if available. Tumor specimens may be collected at baseline and any time after completion of C4 per DI discretion by the CT guided biopsies. . Biopsies may be obtained from primary tumor sites and metastatic sites (if applicable). No more than 4 cores may be obtained per site. When possible, the same site of disease should be biopsied on repeat evaluation.

Specimens are used for the following studies conducted in the LTIB, CCR, NCI:

5.1.1.2.1 Level of PD-L1 expression will be assessed by immunohistochemistry staining (IHC). Of note, further techniques to evaluate the expression of PD-L1 and/or marker candidates impacting the targeting or contributing to improve its expression may be also investigated if needed.

5.1.1.2.2 Frequency and localization of tumor-infiltrated leukocytes (e.g., CD8, CD4 T-cells, Treg, NK cells, macrophage (M1/2 profile) by IHC

5.1.1.2.3 Gene expression-based subtyping

Analysis to be performed in an outside lab, to be determined, using the methods described in Guinney, et al.[75]. We will evaluate the clinical and immunologic effects of treatment in the standard and combination arms by subtype. Additional analysis may be performed to evaluate the expression of immune-related genes in tumor tissue pre- and post-treatment in each arm.

5.1.1.3 Sample Processing

5.1.1.3.1 Blood samples will be processed at:

Clinical Services Program
NCI Frederick Cancer Research and Development Center
PO Box B
Frederick, MD 21702
301-846-1000

On days samples are drawn, Jennifer Bangh at CSP should be notified (phone: [301] 846-5893; fax [301] 846-6222). She will arrange courier delivery of the specimens to the processing lab. Couriers are assigned to CCR and Georgetown and this mechanism is in place.

The weekly patient lists of samples drawn will be emailed to Caroline Jochems at jochemscm@mail.nih.gov, Jen Bangh at jb478s@nih.gov, and Theresa Burks at burkst@mail.nih.gov.

5.1.1.3.2 Tissue samples will be processed immediately after acquisition by a member of the clinical staff. Core biopsy material will be separated (if necessary) into 3 samples. One sample will be placed in formalin and taken to the Laboratory of Pathology and stored as FFPE (formalin fixed paraffin embedded) for IHC examination at a later time. The second sample will be placed in a 1-2mL cryovial and snap-frozen in liquid nitrogen. The third sample will be placed in OCT media for preservation and stored at –70˚C. Any additional samples, if available, will be flash frozen.

After processing, all non-FFPE samples will be transferred for labeling and tracking at:

Clinical Services Program
NCI Frederick Cancer Research and Development Center
PO Box B
Frederick, MD 21702
301-846-1000

On days samples are drawn, Jennifer Bangh at CSP should be notified (phone: [301] 846-5893; fax [301] 846-6222). She will arrange courier delivery of the specimens to the processing lab. Couriers are assigned to CCR and Georgetown and this mechanism is in place.

The weekly patient lists of samples drawn will be emailed to Caroline Jochems at jochemscm@mail.nih.gov, Jen Bangh at jb478s@nih.gov and Theresa Burks (burkst@mailnih.gov).

5.1.2 Omics Studies

Coded, linked, formalin-fixed, paraffin-embedded tissue and blood samples will be sent to NantOmics per the Genomic and Proteomic Sample Collection Manual under an existing CRADA

agreement for analysis of genetic mutations, RNA expression profiling, and proteomic profiling of patient tumor samples from baseline and on treatment.

5.1.2.1 Rationale

Genomic sequencing of tumor cells relative to non-tumor cells from whole blood will be profiled to identify the genomic variances that may contribute to response or disease progression and provide an understanding of molecular abnormalities. RNA sequencing will be conducted to provide expression data and give relevance to DNA mutations. Quantitative proteomics will be conducted to determine the exact amounts of specific proteins and to confirm expression of genes that are correlative of response and disease progression. All genomic, transcriptomic, and proteomic molecular analysis will be retrospective and exploratory, and used for future hypothesis generation.

5.1.2.2 Sampling

Please follow the instructions in the Genomics and Proteomic Sample Collection Manual to become familiar with the materials provided, to register and to create a patient order. For additional questions, please contact the study coordinator as listed on the title page.

Exploratory genomics and proteomics molecular profiling will be performed on formalin-fixed, paraffin embedded (FFPE) tumor tissue and whole blood (subject matched normal comparator against the tumor tissue) by next-generation sequencing and mass spectrometry-based quantitative proteomics. Collection of whole blood at baseline is mandatory for this study. Tumor samples may be collected at baseline and any time after completion of C4 per PI discretion if the patient is willing.

5.1.2.2.1 General Information for Collection of Tumor Tissues and Whole Blood

Tumor tissue and whole blood samples are to be collected according to the instruction cards included in the Tissue Specimen Kit and Blood Specimen Kit. The kits include the materials necessary to collect and ship FFPE tumor tissue samples and whole blood samples.

The Investigational Site is responsible for obtaining the subject's written Informed Consent Form for genomic and proteomic molecular profiling; the Investigator is to confirm that the written informed consent was obtained before collection of genomic and proteomic samples.

The Investigational Site is responsible for completing the P152525 Clinical Trial Requisition Form with the following information:

- Clinical Site Information
- Patient Information
- Specimen Information
- Specimen Collection Information

The Investigational Site will return the completed Clinical Trial Requisition Form via Fax per the instructions provided on the form.

Detailed specimen requirements and procedural instructions for FFPE tumor tissue samples and whole blood samples are provided in the P152525 Genomic and Proteomic Sample Collection Manual.

5.1.2.3 Genomic and Proteomic Analysis of Tumor Tissue and Whole Blood

A single FFPE tumor tissue block is required for the extraction of tumor DNA, tumor RNA, and tumor protein. A whole blood sample is required for the extraction of subject normal DNA. Tumor tissue and whole blood will be processed in the NantOmics, LLC CLIA-registered and CAP accredited/CLIA certified laboratories.

5.1.2.4 Genomic and Proteomic Analysis Results

The individual subject's summary report of cancer-related genomic and proteomic data can be provided to the investigator upon request after the individual subject has completed or discontinued the clinical study. Procedural instructions for requesting the cancer-related genomic and proteomic summary report are provided in the P152525 Genomic and Proteomic Sample Collection Manual.

5.2 SAMPLE COLLECTION SUMMARY

Test/assay	**Sample/ volume blood**	**Type of tube**	**Collection point (+/- 48hrs)**	**Location of specimen analysis**
Tissue analysis (**5.1.1.2**)	Optional core biopsy	N/A	Baseline (from optional fresh or archival samples collected); any time after completion of C4 per DI discretion	Clinical Services Program, Frederick
Omics studies (**5.1.2**)	Single block FFPE with tissue surface area 25 mm2, 75 µm thick, at least 30% malignant tissue	N/A		NantOmics
	Optional biopsy	N/A	leftover samples from biopsies collected on study will be utilized	NantOmics

Test/assay	Sample/ volume blood	Type of tube	Collection point (+/- 48hrs)	Location of specimen analysis
	2. 5 mL blood	PAXgene Blood DNA Tube (provided in kit)	Baseline	NantOmics
CEA-specific T cell response (**5.1.1.1.2**)	15 mL blood	Two 10 mL Sodium heparin	C1D1, C3D1, C5D1, C9D1 and every 4 cycles thereafter	Clinical Services Program, Frederick
PMBC subset correlation (**5.1.1.1.1**)	15 mL blood	Two 10 mL Sodium heparin	C1D1, C3D1, C5D1, C9D1 and every 4 cycles thereafter	Clinical Services Program, Frederick
T cell clonality (**5.1.1.1.4**)	4 mL blood	One 10 mL Sodium heparin	C1D1, C3D1, C5D1, C9D1 and every 4 cycles thereafter	Clinical Services Program, Frederick
Cytokine analysis (**5.1.1.1.3**, **5.1.1.1.5**)	8 mL blood	One SST	C1D1, C3D1, C5D1, C9D1 and every 4 cycles thereafter	Clinical Services Program, Frederick

5.3 SAMPLE STORAGE, TRACKING, DISPOSITION (CLINICAL SERVICES PROGRAM [CSP])

Samples collected at NIH will be ordered in CRIS and tracked through a Clinical Trial Data Management system. Should a CRIS screen not be available, the CRIS downtime procedures will be followed. At participating sites, samples will be ordered and tracked per local SOPs. Samples will not be sent outside NIH without IRB notification and an executed MTA.

All data associated with the patient samples is protected by using a secure database. All samples drawn at the NIH Clinical Center and the Georgetown Lombardi Cancer Center will be transported to the NCI Frederick Central Repository by couriers.

Samples will be tracked and managed by the Central Repository database. All samples will be stored in liquid nitrogen. These freezers are located at NCI Frederick Central Repository in Frederick, Maryland.

NCI Frederick Central Repositories store, among other things, biological specimens in support of NIH clinical studies. All specimens are stored in secure, limited access facilities with sufficient security, back-up and emergency support capability and monitoring to ensure long-term integrity of the specimens for research.

It is the intent and purpose of ATCC to accept only coded, linked, samples and sample information. To the best of our ability, every effort will be made to ensure that protected information is not sent electronically or by hard copy or on vial labels.

Sample data is stored in the BioSpecimen Inventory (BSI) System II. This inventory tracking system is used to manage the storage and retrieval of specimens as well as maintain specimen data. BSI is designed for controlled, concurrent access. It provides a real-time, multi-user environment for tracking millions of specimens. The system controls how and in what order database updates and searches are performed. This control prevents deadlocks and race conditions. For security, BSI has user password access, 3 types of user access levels, and 36 user permissions (levels of access) that can be set to control access to the system functions. BSI provides audit tracking for processes that are done to specimens including shipping, returning to inventory, aliquoting, thawing, additives, and other processes. BSI tracks the ancestry of specimens as they are aliquoted, as well as discrepancies and discrepancy resolution for specimens received by the repository. If a specimen goes out of the inventory, the system maintains data associated with the withdrawal request. Vials are labeled with a unique BSI ID which is printed in both eye-readable and bar-coded format. No patient-specific information is encoded in this ID.

Investigators are granted view, input and withdrawal authority only for their specimens. They may not view specimen data or access specimens for which they have not been authorized. Access to specimen storage is confined to repository staff. Visitors to the repositories are escorted by repository staff at all times.

5.3.1 Protocol Completion/Sample Destruction

All specimens obtained in the protocol are used as defined in the protocol. Any specimens remaining at the completion of the protocol will be stored in the conditions described above. The study will remain open as long as sample or data analysis continues. Samples from consenting subjects will be stored until they are no longer of scientific value or until a subject withdraws consent for their continued use, at which time they will be destroyed. Once primary research objectives for the protocol are achieved, intramural researchers can request access to remaining samples, provided they have an IRB-approved protocol and patient consent or an exemption from OHSRP. The PI will record any loss or unanticipated destruction of samples as a deviation. Reporting will be per the requirements of section Error! Reference source not found..

5.4 SAMPLES FOR GENETIC/GENOMIC ANALYSIS

5.4.1 Whole Genome / RNA expression / Proteomic Analysis

Please see section **5.1.2**.

5.4.2 Certificate of Confidentiality

As part of study efforts to provide confidentiality of subject information, this study has obtained a Certificate of Confidentiality which helps to protect personally identifiable research information.

The Certificate of Confidentiality allows investigators on this trial to refuse to disclose identifying information related to the research participants, should such disclosure have adverse consequences for subjects or damage their financial standing, employability, insurability or reputation. The informed consent includes the appropriate coverage and restrictions of the Certificate of Confidentiality.

5.4.3 Management of Results

Subjects at NIH and at participating sites will not be contacted with information about their gene variant changes because NantOmics will provide the individual subject's report of genomic data to the investigator only after subject has completed or discontinued the clinical study.

6 DATA COLLECTION AND EVALUATION

6.1 DATA COLLECTION

Data will be entered in C3D at all study sites. All data will be kept secure. The PI will be responsible for overseeing entry of data into an in-house password protected electronic system and ensuring data accuracy, consistency and timeliness. The principal investigator, associate investigators/research nurses and/or a contracted data manager will assist with the data management efforts. All human subjects personally identifiable information (PII) as defined in accordance to the Health Insurance Portability and Accountability Act, eligibility and consent verification will be recorded. Primary data obtained during the conduct of the protocol will be kept in secure network drives or in approved alternative sites that comply with NIH security standards. Primary and final analyzed data will have identifiers so that research data can be attributed to an individual human subject participant.

All adverse events, including clinically significant abnormal findings on laboratory evaluations, regardless of severity, will be followed until return to baseline or stabilization of event.

Document AEs from the first study intervention, Study Day 1 through 30 days after removal from study treatment or until off-study, whichever comes first. Adverse events that are serious need to be recorded through 30 days after the last intervention. Beyond 30 days after the last intervention and through long term follow up (survival of subject), only adverse events which are serious and related to the study intervention need to be recorded.

An abnormal laboratory value will be recorded in the database as an AE **only** if the laboratory abnormality is characterized by any of the following:

- Results in discontinuation from the study
- Is associated with clinical signs or symptoms
- Requires treatment or any other therapeutic intervention
- Is associated with death or another serious adverse event, including hospitalization.
- Is judged by the Investigator to be of significant clinical impact
- If any abnormal laboratory result is considered clinically significant, the investigator will provide details about the action taken with respect to the test drug and about the patient's outcome.

Exceptions to data collection: Grade 1 adverse events will not be recorded.

End of study procedures: Data will be stored according to HHS, FDA regulations and NIH Intramural Records Retention Schedule as applicable.

Loss or destruction of data: Should we become aware that a major breach in our plan to protect subject confidentiality and trial data has occurred, this will be reported expeditiously per requirements in section Error! Reference source not found...

6.2 DATA SHARING PLANS

6.2.1 Human Data Sharing Plan

What data will be shared?

I will share human data generated in this research for future research as follows:

_**X**_ Coded, linked data in an NIH-funded or approved public repository.

_**X**_ Coded, linked data in BTRIS (automatic for activities in the Clinical Center)

_**X**_ Identified or coded, linked data with approved outside collaborators under appropriate agreements.

How and where will the data be shared?

Data will be shared through:

_**X**_ An NIH-funded or approved public repository clinicaltrials.gov.

_**X**_ BTRIS (automatic for activities in the Clinical Center)

_**X**_ Approved outside collaborators under appropriate individual agreements.

_**X**_ Publication and/or public presentations.

When will the data be shared?

_**X**_ Before publication.

_**X**_ At the time of publication or shortly thereafter.

6.3 GENOMIC DATA SHARING PLAN

Unlinked genomic data will be deposited in public genomic databases such as dbGaP in compliance with the NIH Genomic Data Sharing Policy.

6.4 RESPONSE CRITERIA

For the purposes of this study, patients should be re-evaluated for response every 8 weeks. In addition to a baseline scan, confirmatory scans should also be obtained up to 6 (not less than 4) weeks following initial documentation of objective response.

Response and progression will be evaluated in this study using the new international criteria proposed by the revised Response Evaluation Criteria in Solid Tumors (RECIST) guideline

(version 1.1).[76] Changes in the largest diameter (unidimensional measurement) of the tumor lesions and the shortest diameter in the case of malignant lymph nodes are used in the RECIST criteria.

6.4.1 Definitions

Evaluable for toxicity: All patients will be evaluable for toxicity from the time of their first treatment with avelumab.

Evaluable for DLT: The first 6 subjects enrolled will be evaluable for DLT from the time of their first treatment with avelumab until 28 days have passed. Subjects who do not complete the DLT observation period for reasons other than a DLT will be replaced.

Evaluable for objective response: Only those patients who have measurable disease present at baseline, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for response. These patients will have their response classified according to the definitions stated below.

Evaluable Non-Target Disease Response: Patients who have lesions present at baseline that are evaluable but do not meet the definitions of measurable disease, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for non-target disease. The response assessment is based on the presence, absence, or unequivocal progression of the lesions.

6.4.2 Disease Parameters

Measurable disease: Measurable lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as:

- By chest x-ray: ≥20 mm;
- By CT scan:
    - Scan slice thickness 5 mm or under as ≥10 mm with CT scan
    - Scan slice thickness >5 mm: double the slice thickness
- With calipers on clinical exam: ≥10 mm.

All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Malignant lymph nodes. To be considered pathologically enlarged and measurable, a lymph node must be ≥15 mm in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm). At baseline and in follow-up, only the short axis will be measured and followed.

Non-measurable disease. All other lesions (or sites of disease), including small lesions (longest diameter <10 mm or pathological lymph nodes with ≥10 to <15 mm short axis), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonitis, inflammatory breast disease, and abdominal masses (not followed by CT or MRI), are considered as non-measurable.

Note: Cystic lesions that meet the criteria for radiographically defined simple cysts should not be considered as malignant lesions (neither measurable nor non-measurable) since they are, by definition, simple cysts.

'Cystic lesions' thought to represent cystic metastases can be considered as measurable lesions, if they meet the definition of measurability described above. However, if non-cystic lesions are present in the same patient, these are preferred for selection as target lesions.

Target lesions. All measurable lesions up to a maximum of 2 lesions per organ and 5 lesions in total, representative of all involved organs, should be identified as **target lesions** and recorded and measured at baseline. Target lesions should be selected on the basis of their size (lesions with the longest diameter), be representative of all involved organs, but in addition should be those that lend themselves to reproducible repeated measurements. It may be the case that, on occasion, the largest lesion does not lend itself to reproducible measurement in which circumstance the next largest lesion which can be measured reproducibly should be selected. A sum of the diameters (longest for non-nodal lesions, short axis for nodal lesions) for all target lesions will be calculated and reported as the baseline sum diameters. If lymph nodes are to be included in the sum, then only the short axis is added into the sum. The baseline sum diameters will be used as reference to further characterize any objective tumor regression in the measurable dimension of the disease.

Non-target lesions. All other lesions (or sites of disease) including any measurable lesions over and above the 5 target lesions should be identified as **non-target lesions** and should also be recorded at baseline. Measurements of these lesions are not required, but the presence, absence, or in rare cases unequivocal progression of each should be noted throughout follow-up.

6.4.3 Methods for Evaluation of Measurable Disease

All measurements should be taken and recorded in metric notation using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation is preferred to evaluation by clinical examination unless the lesion(s) being followed cannot be imaged but are assessable by clinical exam.

Clinical lesions: Clinical lesions will only be considered measurable when they are superficial (e.g., skin nodules and palpable lymph nodes) and ≥10 mm diameter as assessed using calipers (e.g., skin nodules). In the case of skin lesions, documentation by color photography, including a ruler to estimate the size of the lesion, is recommended.

Chest x-ray: Lesions on chest x-ray are acceptable as measurable lesions when they are clearly defined and surrounded by aerated lung. However, CT is preferable.

Conventional CT and MRI: This guideline has defined measurability of lesions on CT scan based on the assumption that CT slice thickness is 5 mm or less. If CT scans have slice thickness greater than 5 mm, the minimum size for a measurable lesion should be twice the slice thickness. MRI is also acceptable in certain situations (e.g. for body scans).

Use of MRI remains a complex issue. MRI has excellent contrast, spatial, and temporal resolution; however, there are many image acquisition variables involved in MRI, which greatly impact image quality, lesion conspicuity, and measurement. Furthermore, the availability of MRI is variable globally. As with CT, if an MRI is performed, the technical specifications of the scanning sequences used should be optimized for the evaluation of the type and site of disease. Furthermore, as with CT, the modality used at follow-up should be the same as was used at baseline and the

lesions should be measured/assessed on the same pulse sequence. It is beyond the scope of the RECIST guidelines to prescribe specific MRI pulse sequence parameters for all scanners, body parts, and diseases. Ideally, the same type of scanner should be used and the image acquisition protocol should be followed as closely as possible to prior scans. Body scans should be performed with breath-hold scanning techniques, if possible.

PET-CT: At present, the low dose or attenuation correction CT portion of a combined PET-CT is not always of optimal diagnostic CT quality for use with RECIST measurements. However, if the site can document that the CT performed as part of a PET-CT is of identical diagnostic quality to a diagnostic CT (with IV and oral contrast), then the CT portion of the PET-CT can be used for RECIST measurements and can be used interchangeably with conventional CT in accurately measuring cancer lesions over time. Note, however, that the PET portion of the CT introduces additional data which may bias an investigator if it is not routinely or serially performed.

Ultrasound: Ultrasound is not useful in assessment of lesion size and should not be used as a method of measurement. Ultrasound examinations cannot be reproduced in their entirety for independent review at a later date and, because they are operator dependent, it cannot be guaranteed that the same technique and measurements will be taken from one assessment to the next. If new lesions are identified by ultrasound in the course of the study, confirmation by CT or MRI is advised. If there is concern about radiation exposure at CT, MRI may be used instead of CT in selected instances.

Endoscopy, Laparoscopy: The utilization of these techniques for objective tumor evaluation is not advised. However, such techniques may be useful to confirm complete pathological response when biopsies are obtained or to determine relapse in trials where recurrence following complete response (CR) or surgical resection is an endpoint.

Tumor markers: Tumor markers alone cannot be used to assess response. If markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response. Specific guidelines for both CA-125 response (in recurrent ovarian cancer) and PSA response (in recurrent prostate cancer) have been published[77-79]. In addition, the Gynecologic Cancer Intergroup has developed CA-125 progression criteria which are to be integrated with objective tumor assessment for use in first-line trials in ovarian cancer[80].

Cytology, Histology: These techniques can be used to differentiate between partial responses (PR) and complete responses (CR) in rare cases (e.g., residual lesions in tumor types, such as germ cell tumors, where known residual benign tumors can remain).

The cytological confirmation of the neoplastic origin of any effusion that appears or worsens during treatment when the measurable tumor has met criteria for response or stable disease is mandatory to differentiate between response or stable disease (an effusion may be a side effect of the treatment) and progressive disease.

FDG-PET: While FDG-PET response assessments need additional study, it is sometimes reasonable to incorporate the use of FDG-PET scanning to complement CT scanning in assessment of progression (particularly possible 'new' disease). New lesions on the basis of FDG-PET imaging can be identified according to the following algorithm:

a. Negative FDG-PET at baseline, with a positive FDG-PET at follow-up is a sign of PD based on a new lesion.

b. No FDG-PET at baseline and a positive FDG-PET at follow-up: If the positive FDG-PET at follow-up corresponds to a new site of disease confirmed by CT, this is PD. If the positive FDG-PET at follow-up is not confirmed as a new site of disease on CT, additional follow-up CT scans are needed to determine if there is truly progression occurring at that site (if so, the date of PD will be the date of the initial abnormal FDG-PET scan). If the positive FDG-PET at follow-up corresponds to a pre-existing site of disease on CT that is not progressing on the basis of the anatomic images, this is not PD.
c. FDG-PET may be used to upgrade a response to a CR in a manner similar to a biopsy in cases where a residual radiographic abnormality is thought to represent fibrosis or scarring. The use of FDG-PET in this circumstance should be prospectively described in the protocol and supported by disease-specific medical literature for the indication. However, it must be acknowledged that both approaches may lead to false positive CR due to limitations of FDG-PET and biopsy resolution/sensitivity.

Note: A 'positive' FDG-PET scan lesion means one which is FDG avid with an uptake greater than twice that of the surrounding tissue on the attenuation corrected image.

6.4.4 Response Criteria

6.4.4.1 Evaluation of Target Lesions

Complete Response (CR): Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.

Partial Response (PR): At least a 30% decrease in the sum of the diameters of target lesions, taking as reference the baseline sum of diameters.

Progressive Disease (PD): At least a 20% increase in the sum of the diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progressions).

Stable Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum of diameters while on study.

6.4.4.2 Evaluation of Non-Target Lesions

Complete Response (CR): Disappearance of all non-target lesions and normalization of tumor marker level. All lymph nodes must be non-pathological in size (<10 mm short axis).

Note: If tumor markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response.

Non-CR/Non-PD: Persistence of one or more non-target lesion(s) and/or maintenance of tumor marker level above the normal limits.

Progressive Disease (PD): Appearance of one or more new lesions and/or *unequivocal progression* of existing non-target lesions. *Unequivocal progression* should not normally trump target lesion status. It must be representative of overall disease status change, not a single lesion increase.

Although a clear progression of "non-target" lesions only is exceptional, the opinion of the treating physician should prevail in such circumstances, and the progression status should be confirmed at a later time by the review panel (or Principal Investigator).

6.4.4.3 Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria.

For Patients with Measurable Disease (i.e., Target Disease)

Target Lesions	Non-Target Lesions	New Lesions	Overall Response	Best Overall Response when Confirmation is Required*
CR	CR	No	CR	≥4 wks. Confirmation**
CR	Non-CR/Non-PD	No	PR	≥4 wks. Confirmation**
CR	Not evaluated	No	PR	
PR	Non-CR/Non-PD/not evaluated	No	PR	
SD	Non-CR/Non-PD/not evaluated	No	SD	Documented at least once ≥4 wks. from baseline**
PD	Any	Yes or No	PD	no prior SD, PR or CR
Any	PD***	Yes or No	PD	
Any	Any	Yes	PD	

* See RECIST 1.1 manuscript for further details on what is evidence of a new lesion.

** Only for non-randomized trials with response as primary endpoint.

*** In exceptional circumstances, unequivocal progression in non-target lesions may be accepted as disease progression.

Note: Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be reported as "*symptomatic*

deterioration." Every effort should be made to document the objective progression even after discontinuation of treatment.

For Patients with Non-Measurable Disease (i.e., Non-Target Disease)

Non-Target Lesions	New Lesions	Overall Response
CR	No	CR
Non-CR/non-PD	No	Non-CR/non-PD*
Not all evaluated	No	not evaluated
Unequivocal PD	Yes or No	PD
Any	Yes	PD

* 'Non-CR/non-PD' is preferred over 'stable disease' for non-target disease since SD is increasingly used as an endpoint for assessment of efficacy in some trials so to assign this category when no lesions can be measured is not advised

6.4.5 Duration of Response

Duration of overall response: The duration of overall response is measured from the time measurement criteria are met for CR or PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since the treatment started).

The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that progressive disease is objectively documented.

Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started, including the baseline measurements.

6.4.6 Progression-Free Survival (PFS)

PFS is defined as the duration of time from start of treatment to time of progression or death, whichever occurs first.

6.4.7 Response Review

An independent review of CT scans and MRIs used in determining response will be undertaken at the end of the study by a collaborator in the Radiology and Imaging Sciences. Images will be reviewed using RECIST 1.1. Initial evaluation of progression should be confirmed with a follow up scan in 4-6 weeks, if clinically reasonable. Date of progression should be the initial date of progression when confirmation is obtained. If no confirmation scan is performed, the initial scan demonstrating progression will determine progression.

The image data which is collected from the sites outside the NIH LAN, will be using the NIH provided secure e-mail system or FEDEX of a CD with images. The system provides the ability to

e-mail large attachments which would include the dicom images collected at the remote sites collaborating with the clinical trial.

When a remote site has an image data set which is ready to be transferred to the NCI site for central storage, they should send a non-secure e-mail to a designated e-mail recipient in the NCI informing them that data is ready to be transferred. The recipient of that e-mail will send an NIH secure e-mail to the offsite person requesting that they send the data. The offsite person will then, by replying to the NIH secure e-mail, attach the DICOM image data set to the replied too e-mail. The NCI person will receive the replied too e-mail with the data attached. The NCI person will extract the attachment and upload the offset dicom image data set to the NCI PACS for final storage and archival.

Alternatively, the images can be loaded to a CD and FEDEX to the NCI designated person.

NCI designated recipients:

Chrisa Thomas
10 Center Drive, Building 10
Room 13N210
Bethesda, MD 20892.

6.5 TOXICITY CRITERIA

The following adverse event management guidelines are intended to ensure the safety of each patient while on the study. The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_40).

7 NIHREPORTING REQUIREMENTS/DATA AND SAFETY MONITORING PLAN

7.1 DEFINITIONS

Please refer to definitions provided in Policy 801: Reporting Research Events found here.

7.2 OHSRP OFFICE OF COMPLIANCE AND TRAINING IRB REPORTING

7.2.1 Expedited Reporting

Please refer to the reporting requirements in Policy 801: Reporting Research Events and Policy 802 Non-Compliance Human Subjects Research found here. Note: Only IND Safety Reports that meet the definition of an unanticipated problem will need to be reported per these policies.

7.2.2 IRB Requirements for PI Reporting at Continuing Review

Please refer to the reporting requirements in Policy 801: Reporting Research Events found here.

7.3 NCI CLINICAL DIRECTOR REPORTING

Problems expeditiously reported to the OHSRP in iRIS will also be reported to the NCI Clinical Director. A separate submission is not necessary as reports in iRIS will be available to the Clinical Director.

In addition to those reports, all deaths that occur within 30 days after receiving a research intervention should be reported via email to the Clinical Director unless they are due to progressive disease.

To report these deaths, please send an email describing the circumstances of the death to Dr. Dahut at NCICCRQA@mail.nih.gov within one business day of learning of the death

7.4 .NCI GUIDANCE FOR REPORTING EXPEDITED ADVERSE EVENTS FOR MULTI-CENTER TRIALS

Please refer to the reporting requirements in Policy 801: Reporting Research Events and Policy 802: Non-compliance in Human Subjects Research, found here. Until such time as direct electronic reporting mechanisms are available to participating sites, the site PI must immediately report to the coordinating center PI any deaths possibly related to the research within 24 hours of PI awareness of the event. The Site PI must also report any other events required by Policy 801 to the coordinating center PI within 7 days of PI awareness using the CCR Problem Report Form (See **Appendix C**).

Once direct electronic reporting mechanisms are available, these will be utilized. Please also notify the coordinating center PI and study coordinator of your submission at the time you make it.

For IND studies, the site PI will also directly submit reports to the CCR as IND sponsor per section **8.3.**

7.5 INSTITUTIONAL BIOSAFETY COMMITTEE (IBC) REPORTING CRITERIA

7.5.1 Serious Adverse Event Reports to IBC

The Principal Investigator (or delegate) will notify IBC of any unexpected fatal or life-threatening experience associated with the use of Ad-CEA vaccine as soon as possible but in no event later than 7 calendar days of initial receipt of the information. Serious adverse events that are unexpected and associated with the use of Ad-CEA vaccine, but are not fatal or life-threatening, much be reported to the NIH IBC as soon as possible, but not later than 15 calendar days after the investigator's initial receipt of the information. Adverse events may be reported by using the FDA Form 3500a.

7.5.2 Annual Reports to IBC

Within 60 days after the one-year anniversary of the date on which the IBC approved the initial protocol, and after each subsequent anniversary until the trial is completed, the Principal Investigator (or delegate) shall submit the information described below. Alternatively, the IRB continuing review report can be sent to the IBC in lieu of a separate report. Please include the IBC protocol number on the report.

7.5.2.1 Clinical Trial Information

A brief summary of the status of the trial in progress or completed during the previous year. The summary is required to include the following information:

- the title and purpose of the trial;
- clinical site;
- the Principal Investigator;
- clinical protocol identifiers;
- participant population (such as disease indication and general age group, e.g., adult or pediatric);
- the total number of participants planned for inclusion in the trial; the number entered into the trial to date whose participation in the trial was completed; and the number who dropped out of the trial with a brief description of the reasons;
- the status of the trial, e.g., open to accrual of subjects, closed but data collection ongoing, or fully completed;
- if the trial has been completed, a brief description of any study results.

7.5.2.2 Progress Report and Data Analysis

Information obtained during the previous year's clinical and non-clinical investigations, including:

- a narrative or tabular summary showing the most frequent and most serious adverse experiences by body system;
- a summary of all serious adverse events submitted during the past year;
- a summary of serious adverse events that were expected or considered to have causes not associated with the use of the gene transfer product such as disease progression or concurrent medications;
- if any deaths have occurred, the number of participants who died during participation in the investigation and causes of death;
- a brief description of any information obtained that is pertinent to an understanding of the gene transfer product's actions, including, for example, information about dose-response, information from controlled trials, and information about bioavailability.

7.6 NIH REQUIRED DATA AND SAFETY MONITORING PLAN

7.6.1 Principal Investigator/Research Team

The clinical research team will meet on a regular basis (every two weeks by teleconference with participating sites) when patients are being actively treated on the trial to discuss each patient, enrollment and data management issues. Decisions about dose level enrollment and dose escalation (NCI site only) will be made based on the toxicity data from prior patients.

All data will be collected in a timely manner and reviewed by the principal investigator or a lead associate investigator. Events meeting requirements for expedited reporting as described in section Error! Reference source not found. will be submitted within the appropriate timelines.

The principal investigator will review adverse event and response data on each patient to ensure safety and data accuracy. The principal investigator will personally conduct or supervise the investigation and provide appropriate delegation of responsibilities to other members of the research staff.

7.6.2 Safety Monitoring Committee (SMC)

This protocol will require oversight from the Safety Monitoring Committee (SMC). Initial review will occur as soon as possible after the annual NIH Intramural IRB continuing review date. Subsequently, each protocol will be reviewed as close to annually as the quarterly meeting schedule permits or more frequently as may be required by the SMC. For initial and subsequent reviews, protocols will not be reviewed if there is no accrual within the review period. Written outcome letters will be generated in response to the monitoring activities and submitted to the Principal investigator and Clinical Director or Deputy Clinical Director, CCR, NCI.

8 SPONSOR SAFETY REPORTING

8.1 DEFINITIONS

8.1.1 Adverse Event

Any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have a causal relationship with this treatment. An adverse event (AE) can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product (ICH E6 (R2))

8.1.2 Serious Adverse Event (SAE)

An adverse event or suspected adverse reaction is considered serious if in the view of the investigator or the sponsor, it results in any of the following:

- Death,
- A life-threatening adverse event (see section **8.1.3**)
- Inpatient hospitalization or prolongation of existing hospitalization
    - A hospitalization/admission that is pre-planned (i.e., elective or scheduled surgery arranged prior to the start of the study), a planned hospitalization for pre-existing condition, or a procedure required by the protocol, without a serious deterioration in health, is not considered a serious adverse event.
    - A hospitalization/admission that is solely driven by non-medical reasons (e.g., hospitalization for patient convenience) is not considered a serious adverse event.

- Emergency room visits or stays in observation units that do not result in admission to the hospital would not be considered a serious adverse event. The reason for seeking medical care should be evaluated for meeting one of the other serious criteria.

- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

8.1.3 Life-threatening

An adverse event or suspected adverse reaction is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death. (21CFR312.32)

8.1.4 Severity

The severity of each Adverse Event will be assessed utilizing the CTCAE version 5.0.

8.1.5 Relationship to Study Product

All AEs will have their relationship to study product assessed using the terms: related or not related.

- Related – There is a reasonable possibility that the study product caused the adverse event. Reasonable possibility means that there is evidence to suggest a causal relationship between the study product and the adverse event.

- Not Related – There is not a reasonable possibility that the administration of the study product caused the event.

8.2 Assessment of Safety Events

AE information collected will include event description, date of onset, assessment of severity and relationship to study product and alternate etiology (if not related to study product), date of resolution of the event, seriousness and outcome. The assessment of severity and relationship to the study product will be done only by those with the training and authority to make a diagnosis and listed on the Form FDA 1572 as the site principal investigator or sub-investigator. AEs occurring during the collection and reporting period will be documented appropriately regardless of relationship. AEs will be followed through resolution.

SAEs will be:

- Assessed for severity and relationship to study product and alternate etiology (if not related to study product) by a licensed study physician listed on the Form FDA 1572 as the site principal investigator or sub-investigator.

- Recorded on the appropriate SAE report form, the medical record and captured in the clinical database.
- Followed through resolution by a licensed study physician listed on the Form FDA 1572 as the site principal investigator or sub-investigator.

For timeframe of recording adverse events, please refer to section Error! Reference source not found.. All serious adverse events recorded from the time of first investigational product administration must be reported to the sponsor with the exception of any listed in section **8.4**.

8.3 Reporting of Serious Adverse Events

Any AE that meets protocol-defined serious criteria or meets the definition of Adverse Event of Special Interest that require expedited reporting must be submitted immediately (within 24 hours of awareness) to OSRO Safety using the CCR SAE report form. Any exceptions to the expedited reporting requirements are found in section **8.4**.

All SAE reporting must include the elements described in section **8.2**.

SAE reports will be submitted to the Center for Cancer Research (CCR) at: OSROSafety@mail.nih.gov and to the CCR PI and study coordinator. CCR SAE report form and instructions can be found at: https://ccrod.cancer.gov/confluence/pages/viewpage.action?pageId=157942842

Following the assessment of the SAE by OSRO, other supporting documentation of the event may be requested by the OSRO Safety and should be provided as soon as possible.

8.4 Waiver of expedited reporting to CCR

As hospitalization due to disease progression is part of the secondary objective, and captured as an endpoint in this study, hospitalization due to disease progression will not be reported in expedited manner to the sponsor. However, if there is evidence suggesting a causal relationship between the study drug and the event, report the event in an expedited manner according to section Error! Reference source not found..

8.5 Safety Reporting Criteria to the Pharmaceutical Collaborators

8.5.1 EMD-Serono (Avelumab)

To be sent by Office of Sponsor and Regulatory Oversight, CCR, NCI/NIH:

The following reportable events must be submitted to EMD Serono within 2 business days or 3 calendar days (whichever comes first) using the applicable safety report form provided. The Sponsor will assume responsibility for submitting the reportable event(s) to EMD Serono as well as ensuring that any local reporting requirements are completed in parallel.

- Serious Adverse Events
- Exposure during Pregnancy or Breastfeeding (even if not associated with an adverse event)
- Occupational exposure (even if not associated with an adverse event)

- Potential drug-induced liver injury (Hy's Law cases): These events are considered important medical events and should be reported as SAEs.

Contact information for submission of reportable events to EMD Serono:

Fax: +49 6151 72 6914

OR

E-mail: GlobalDrugSafety@merckgroup.com

Specifying:

PROTOCOL Number and/or Title

EMD Serono assigned Study Number

SUBJECT Number

SITE Number/PI Name

SAE/ONSET DATE.

8.5.2 Etubics Corporation (Ad-CEA Vaccine)

In the event of any new SAE (of any Grade) occurring during the reporting period, the Manufacturer (Etubics Corporation) or designee must immediately (i.e., within a maximum 24 hours after becoming aware of the event) be informed by telephone, by fax, or by email.

When an event (or follow-up information) is reported by telephone, a written report must be sent immediately thereafter by fax or email.

Reporting procedures and timelines are the same for any new information on a previously reported SAE (follow-up).

Joseph Balint
Address: 410 West Harrison Street, Suite 100, Seattle, WA 98119
Telephone: +1-206-838-5110 ext. 107
Email: joe@etubics.com

OR

Frank Jones
Address: 410 West Harrison Street, Suite 100, Seattle, WA 98119
Telephone: +1-206-838-5110 ext. 101 (office)
+1-206-818-2857 (cell)
Email: frank@etubics.com

All Medwatch 3500a forms and written reports should be transmitted, which must be completed by the investigator following specific completion instructions. Relevant pages from the CRF may be provided in parallel (e.g., medical history, concomitant drugs).

In all cases, the information provided in the SAE Report Form must be consistent with the data on the event that is recorded in the corresponding sections of the CRF.

8.5.3 Reporting Pregnancy

8.5.3.1 Maternal exposure

If a patient becomes pregnant during the course of the study, the study treatment should be discontinued immediately, and the pregnancy reported to the Sponsor no later than 24 hours of when the Investigator becomes aware of it. The Investigator should notify the Sponsor no later than 24 hours of when the outcome of the Pregnancy becomes known,

Pregnancy itself is not regarded as an SAE. However, congenital abnormalities or birth defects and spontaneous miscarriages that meet serious criteria (section **8.1.2**) should be reported as SAEs.

The outcome of all pregnancies (spontaneous miscarriage, elective termination, ectopic pregnancy, normal birth, or congenital abnormality) should be followed up and documented.

8.5.3.2 Paternal exposure

Male patients should refrain from fathering a child or donating sperm during the study and for 4 months after the last treatment with avelumab or 6 months after the last administration of bevacizumab, whichever occurs later.

Pregnancy of the patient's partner is not considered to be an AE. However, the outcome of all pregnancies (spontaneous miscarriage, elective termination, ectopic pregnancy, normal birth, or congenital abnormality) occurring from the date of the first dose until 4 months after the last treatment with avelumab or 6 months after the last administration of bevacizumab, whichever occurs later should, if possible, be followed up and documented.

8.6 REGULATORY REPORTING FOR STUDIES CONDUCTED UNDER CCR-SPONSORED IND

Following notification from the investigator, CCR, the IND sponsor, will report any suspected adverse reaction that is both serious and unexpected. CCR will report an AE as a suspected adverse reaction only if there is evidence to suggest a causal relationship between the study product and the adverse event. CCR will notify FDA and all participating investigators (i.e., all investigators to whom the sponsor is providing drug under its INDs or under any investigator's IND) in an IND safety report of potential serious risks from clinical trials or any other source, as soon as possible, in accordance to 21 CFR Part 312.32.

All serious events will be reported to the FDA at least annually in a summary format.

9 CLINICAL MONITORING

As a sponsor for clinical trials, FDA regulations require the CCR to maintain a monitoring program. The CCR's program allows for confirmation of: study data, specifically data that could affect the interpretation of primary and secondary study endpoints; adherence to the protocol, regulations, ICH E6, and SOPs; and human subjects protection. This is done through independent verification of study data with source documentation focusing on:

- Informed consent process
- Eligibility confirmation
- Drug administration and accountability
- Adverse events monitoring
- Response assessment.

The monitoring program also extends to multi-site research when the CCR is the coordinating center.

This trial will be monitored by personnel employed by a CCR contractor. Monitors are qualified by training and experience to monitor the progress of clinical trials. Personnel monitoring this study will not be affiliated in any way with the trial conduct.

10 STATISTICAL CONSIDERATIONS

Following an initial 6 patient safety evaluation, the primary objective of the trial is to determine if there is a difference in progression free survival among patients with metastatic or unresectable colorectal cancer who are treated with standard of care vs. standard of care + anti-PD-L1+ Ad-CEA vaccine monoclonal antibody. A secondary hypothesis (objective) will be to obtain preliminary data on ratio of patients that are hospitalized because of adverse events attributed to disease progression.

Based upon results in the literature, patients who would be eligible to be randomized on this trial would be expected to have an estimated 10-month median progression free survival with standard of care alone. The goal of this study will be to determine if the use of the anti-PD-L1 monoclonal antibody + the Ad-CEA vaccine along with standard of care will result in patients having an increased median progression free survival of 18 months. Kaplan-Meier curves and a two-tailed log-rank test will be the primary analysis methods. Assuming exponential progression free survival curves, the hazard rate for standard of care alone is 0.0693 or approximately a 7% probability of progressing each month when the median progression free survival is 10 months. If we assume that the combination arm will be associated with a median progression free survival of 18 months, this corresponds to a hazard rate of 0.0385 and the resulting hazard ratio for the comparison of the two overall actuarial curves would be 1.80. Following the principals of a phase 2.5 design, to compare these curves and detect a difference with a 0.10 one-tailed log-rank test, a total of 35 evaluable subjects per arm (70 total) will need to be randomized over a one year period and a maximum follow-up of three years, with occurrence of 52 total progressions, in order to have 80% power to compare the curves.

In addition, during the randomized portion of the trial, beginning with the 4th patient on a given arm, if the cumulative fraction of patients at any point who experience a grade 3 or 4 toxicity on either arm is greater than or equal to 1/3, then accrual to the randomized trial will be suspended and will only resume following modifications to the treatment on the appropriate arm, detailed in an amendment created for that purpose. Thus, for example, one patient with a DLT in the first 3 randomized patients on an arm would be permitted, but if there are 2 patients with a DLT in the first 4 on either arm, this would be considered excessive.

It is expected that all 76 patients (6 for safety lead-in plus 70 for phase 2.5) can be accrued onto this trial in three to four years. In order to allow for a number of inevaluable patients, due to screen failures at the other sites, the accrual ceiling will be set at 97 patients.

11 COLLABORATIVE AGREEMENTS

11.1 CRADA

11.1.1 EMD Serono

A CRADA (02666) is in place between the Laboratory of Tumor Immunology and Biology (LTIB), CCR NCI and EMD Serono, the manufacturer of avelumab.

11.1.2 Etubics Corporation

A CRADA (02997) is in place between the Laboratory of Tumor Immunology and Biology (LTIB), CCR NCI and Etubics Corporation, the manufacturer of Ad-CEA vaccine.

11.1.3 NantOmics

The studies referenced in section **5.1.2** will be conducted under a CRADA (03063) agreement between the Laboratory of Tumor Immunology and Biology (LTIB), CCR NCI and NantOmics.

11.2 MTA

11.2.1 EMD Serono

A MTA (41326-16) is in place between the CCR NCI and EMD Serono

11.3 MULTI-INSTITUTIONAL GUIDELINES

11.3.1 IRB Approvals

The PI will provide the NIH Intramural IRB with a copy of the participating institution's approved yearly continuing review. Registration will be halted at any participating institution in which a current continuing approval is not on file at the NIH Intramural IRB.

11.3.2 Amendments and Consents

The CCR PI will provide the NIH Intramural IRB with copies of all amendments, consents and approvals from each participating institution.

12 HUMAN SUBJECTS PROTECTIONS

12.1 RATIONALE FOR SUBJECT SELECTION

Subjects from all racial and ethnic groups are eligible for this trial if they meet the eligibility criteria. Efforts will be made to extend the accrual to a representative population.

12.2 PARTICIPATION OF CHILDREN

The age group for enrollment on this trial is ≥ 18 years of age.

12.3 PARTICIPATION OF SUBJECTS UNABLE TO GIVE CONSENT

Adults unable to give consent are excluded from enrolling in the protocol. However, re-consent may be necessary and there is a possibility, though unlikely, that subjects could become decisionally impaired. For this reason and because there is a prospect of direct benefit from research participation (section **12.4**), all subjects will be offered the opportunity to fill in their wishes for research and care, and assign a substitute decision maker on the "NIH Advance

Directive for Health Care and Medical Research Participation" form so that another person can make decisions about their medical care in the event that they become incapacitated or cognitively impaired during the course of the study. Note: The PI or AI will contact the NIH Ability to Consent Assessment Team (ACAT) for evaluation as needed for the following: an independent assessment of whether an individual has the capacity to provide consent; assistance in identifying and assessing an appropriate surrogate when indicated; and/or an assessment of the capacity to appoint a surrogate. For those subjects that become incapacitated and do not have pre-determined substitute decision maker, the procedures described in NIH HRPP SOP 14E for appointing a surrogate decision maker for adult subjects who are (a) decisionally impaired, and (b) who do not have a legal guardian or durable power of attorney, will be followed.

12.4 Evaluation of Benefits and Risks/Discomforts

Adult subjects, including those that have lost the capacity to consent

Patients will receive evaluation of their disease at the National Cancer Institute's Clinical Center and/or MedStar Georgetown University Hospital (Lombardi Cancer Center). This protocol may or may not benefit an individual, but the results may help the investigators learn more about the disease and develop new treatments for patients with this disease.

Potential adverse reactions attributable to the administration of avelumab and the chemotherapeutic agents utilized in this trial are discussed in section **13**. All care will be taken to minimize side effects, but they can be unpredictable in nature and severity. Patients will be examined and evaluated prior to enrollment. All evaluations to monitor the treatment of patients will be recorded in the patient chart. If patients suffer any physical injury as a result of the participation in this study, immediate medical treatment is available at the Clinical Center, National Cancer Institute, Bethesda, Maryland and/or MedStar Georgetown University Hospital (Lombardi Cancer Center).

In addition, the study may involve up to two CT guided biopsies (optional) collected for research purposes only. Subjects undergoing two optional biopsy collection will be exposed to 1.5 rem. This amount of radiation is below the guideline of 5 rem per year allowed for adult research subjects by the NIH Radiation Safety Committee.

Although no compensation is available, any injury will be evaluated and treated in keeping with the benefits or care to which patients are entitled under applicable regulations. In all publications and presentations resulting from this trial, patients' anonymity will be protected to the maximum extent possible. Authorized personnel from the National Cancer Institute (NCI) and Food and Drug Administration (FDA) or other regulatory authorities may have access to research files in order to verify that patients' rights have been safeguarded. In addition, patient names will be given to the Central Registration to register and verify patients' eligibility.

12.5 Consent and Assent Process and Documentation

The investigational nature and research objectives of this trial, the procedures and treatments involved and their attendant risks and discomforts and benefits, and potential alternative therapies will be carefully explained to the patient, and a signed informed consent document will be obtained by a study investigator prior to entry onto the study.

The PI or associate investigator will meet with the patient to discuss the protocol treatment and alternative options in detail. It will be stated clearly that participation in the research study is voluntary and that participants can withdraw from the study without losing benefits they would otherwise be entitled to. The patient will be encouraged to ask questions, and additional meetings to discuss the treatment options will be arranged as necessary.

If there is an optional biopsy for research in the protocol, then the patient will consent at the time of the procedure. If the patient refuses the optional biopsy at that time, the refusal will be documented in the medical record.

12.5.1 Telephone re-consent procedure

Re-consent on this study may be obtained via telephone according to the following procedure: the informed consent document will be sent to the subject. An explanation of the study will be provided over the telephone after the subject has had the opportunity to read the consent form. The subject will sign and date the informed consent. The original informed consent document will be sent back to the consenting investigator who will sign and date the consent form with the date the consent was obtained via telephone. A fully executed copy will be returned via mail for the subject's records. The informed consent process will be documented in the medical record.

12.5.2 Request for Waiver of Consent for Screening Activities

Prior to the subject signing the consent for this study pre-screening activities listed in section **2.2.1** may be performed.

We request a waiver of consent for these activities as they involve only minimal risk to the subjects. A waiver will not adversely affect the rights and welfare of the subjects given that the activities are only intended to determine suitability for screening for participation in research protocols. These activities could not practicably be carried out without the wavier as central recruiting services, utilized in the NIH Clinical Center, perform pre-screening activities for multiple studies and obtaining consent for each one is beyond their resources. The subjects will be provided with additional pertinent information after participation as they will be informed whether or not they are eligible to sign a consent for additional screening.

13 PHARMACEUTICAL INFORMATION

All commercial agents are used as indicated, therefore these agents will not be included in the IND.

13.1 AVELUMAB (BB IND# 17056)

13.1.1 Source

Avelumab will be supplied by the manufacturer, EMD Serono, Inc., through a Cooperative Research and Development Agreement (CRADA). The Clinical Trial Supplies department of the manufacturer will supply the trial medication of avelumab, which will be distributed to the sites by EMD Serono, the manufacturer of avelumab.

13.1.2 Toxicity

The available safety data for current EMR100070-001 trial are summarized below based on the safety data cut-off date, 05 November 2014. In addition to subjects treated during dose escalation, a total of 480 subjects were enrolled during the dose expansion (NSCLC: 184; metastatic breast cancer: 169; gastric cancer: 47; colorectal cancer: 22; castrate-resistant prostate cancer: 11; ovarian cancer: 37, melanoma: 5; mesothelioma: 1; adrenocortical carcinoma: 1; and urothelial carcinoma: 3), treated with the recommended dose of 10 mg/kg avelumab once every 2 weeks and followed up for at least 4 weeks up to the cut-off date.

For all subjects treated during the dose expansion, the most frequently affected System Organ Classes (with an incidence > 30%) were general disorders and administration site conditions (59.6%), gastrointestinal disorders (57.3%), respiratory, thoracic and mediastinal disorders (39.8%), musculoskeletal and connective tissue disorders (38.8%), and metabolic and nutrition disorders (32.3%).

Most Frequently Reported Treatment-related TEAEs During Dose Expansion (≥ 5% of Subjects)	
Treatment-emergent Adverse Events[a] **Preferred Term (MedDRA)**	**Subjects (Safety Population, N = 480)** **N (%)**
Fatigue	97 (20.2%)
Nausea	62 (12.9%)
Infusion-related reaction	47 (9.8%)
Chills	33 (6.9%)
Diarrhea	33 (6.9%)
Decreased appetite	30 (6.3%)
Pyrexia	27 (5.6%)
Influenza like illness	25 (5.2%)
Arthralgia	24 (5.0%)

MedDRA = Medical Dictionary for Regulatory Activities.

a Only treatment-emergent adverse events started during the on-treatment period are summarized.

Treatment-related TEAEs Grade ≥ 3

Of the Grade ≥ 3 treatment-related TEAEs (59 subjects; 12.3%), the following occurred in more than 2 subjects: fatigue (5 subjects, 1.0%), anemia (5 subjects, 1.0%), infusion-related reaction, lipase increased, and GGT increased (each in 4 subjects, 0.8%).

Of the 59 subjects who had Grade ≥ 3 treatment-related TEAEs, 44 (9.2%) had Grade 3 treatment-related TEAEs, 11 (2.3%) had Grade 4 treatment-related TEAEs, and 4 (0.8%) had Grade 5

treatment-related TEAEs. One Grade 3 event of encephalopathy (Subject 101-0032), which was initially considered as posterior reversible encephalopathy syndrome, was assessed as related to trial medication with an alternative explanation of hypertension.

Serious Adverse Events

Overall, 176 of the 480 subjects (36.7%) treated during the dose expansion had serious TEAEs. Of these, 22 (4.6%) subjects reported dyspnea, which was the most frequent serious TEAE in this group, followed by 19 subjects (4.0%) reporting disease progression, 12 subjects (2.5%) reporting pleural effusion, 11 subjects (2.3%) reporting pneumonia, and 7 subjects (1.5%) reporting anemia. All other serious TEAEs were each reported in less than 1.5% of subjects.

Of the serious TEAEs considered treatment-related by the investigator (31 subjects; 6.5%), the following were reported for 2 or more subjects: infusion-related reaction (4 subjects, 0.8%), pneumonitis (3 subjects, 0.6%), and disease progression, dyspnea, and hypercalcemia (each in 2 subjects, 0.4%).

Deaths

In total, 134 subjects (27.9%) treated during the dose expansion died up to the cut-off date (05 November 2014). Of these, the majority of deaths (101 deaths; 21.0%) were due to disease progression. An additional 8 deaths (1.7%) were due to TEAEs unrelated to trial treatment, 4 deaths (0.8%) were due to TEAEs related to trial treatment, and the reason for 8 deaths (1.7%) was labeled as other. The reason for 13 deaths (2.7%) was unknown at the time of the data cut-off. Of the 134 subjects who died, 53 subjects (11.0%) died within 30 days of the last administration of trial treatment. Among these deaths, 39 (8.1%) were due to disease progression, 7 (1.5%) were due to TEAEs unrelated to trial treatment, 4 (0.8%) were due to TEAEs related to trial treatment, and 3 (0.6%) were due to other reasons. No death of unknown reason was reported in the 30-day period.

Immune-related Adverse Events

As of 05 November 2014, a cumulative review revealed 56 cases of potential immune-related AEs out of 480 subjects (11.7%) treated in the dose expansion part of trial EMR 100070-001 and 4 cases out of 50 subjects (8.0%) treated in the dose escalation part of trial EMR 100070-001. A customized Medical Dictionary for Regulatory Activities (MedDRA) query was used for data retrieval from the clinical database with predefined Preferred Terms of potential immune-related AEs (irAEs).

Of 69 potential irAEs reported, 13 were SAEs (18.8%) and 56 were non-serious AEs (81.1%). In the majority of the cases, there was a plausible temporal association between the event onset and the drug administration. Of these 69 events, 46 events (66.7%) were assessed as treatment related by the investigator and 23 events (33.3%) were assessed as not treatment-related by the investigator.

Twenty-six events were assessed as Grade 1, 29 events as Grade 2, 11 events as Grade 3, 2 events as Grade 4, and 1 event (pneumonitis) as Grade 5 (Please note: 2 more events of autoimmune hepatitis had a fatal outcome; however, they were assessed as Grade 3 with a consequent fatal liver failure).

Infusion-related Reactions

Two suspected unexpected serious adverse reactions (SUSARs; anaphylactic reaction and infusion-related reaction) involving 2 subjects were reported in December 2013 and triggered a cumulative review of serious and non-serious cases of infusion-related reactions / hypersensitivity across the avelumab program. Following evaluation of safety signals, infusion related reactions / hypersensitivity have been classified as a newly identified risk (previously classified as a potential risk) and a mandatory premedication regimen of histamine H1 receptor (H1) blockers plus acetaminophen was implemented for all trial subjects as of 28 January 2014.

As of 05 November 2014, 49 (10.2%) of the 480 subjects in the expansion cohort experienced at least 1 episode of an infusion-related reaction when receiving avelumab monotherapy. Most of the events were Grade 1 (8 subjects, 1.7%) or Grade 2 (36 subjects, 7.5%) in intensity, and Grade 3 (3 subjects, 0.6%) or Grade 4 events (2 subjects, 0.4%) were less frequent. No Grade 5 events were reported. Most of the infusion-related reaction events had an onset after the first (30 subjects, 6.3%) or second (16 subjects, 3.3%) avelumab infusion. In 8 subjects (1.7%), avelumab treatment was discontinued because of infusion-related reaction events.

In addition, 1 subject (2.0%) in the dose escalation cohort reported an infusion-related reaction event (Grade 2).

In addition to the aforementioned 49 subjects, 1 case of Grade 4 cardiac arrest occurred 1.5 hours after the third infusion of avelumab (10 mg/kg). The subject died due to an anoxic brain injury 7 days later; no autopsy was performed.

Starting from 29 January 2014, the Sponsor has implemented a mandatory premedication with H1 blockers plus acetaminophen for all subjects who are to receive avelumab. This premedication procedure was applied to 28 and 440 subjects in the dose escalation and the pooled treatment expansion cohort, respectively. Under this premedication procedure, 33 of 440 subjects (7.5%) in the expansion cohort experienced infusion-related reaction events, with 6 subjects (1.4%) having Grade 1, 26 subjects (5.9%) having Grade 2, and 1 subject (0.2%) having Grade 3 events. No infusion-related reaction events were reported in the 28 subjects in the dose escalation cohort.

13.1.3 Formulation and preparation

Avelumab is formulated as a 10.0 mg/mL solution in single-use glass vials, with a rubber stopper. Packaging and labeling will be in accordance with applicable local regulatory requirements and applicable Good Manufacturing Practice (GMP) guidelines. Avelumab will be packed in boxes containing a suitable number of vials. The information on the medication will be in accordance with approved submission documents.

Avelumab will be shipped in transport cool containers (2°C to 8°C) that are monitored with temperature control devices.

Prior to preparation, the day of usage, avelumab must be allowed to warm to room temperature (15-25°C) for 30 minutes.

The calculated dose of avelumab must be diluted with 0.9% sterile sodium chloride injection to a total volume of 250 mL in an infusion bag. Prior to the addition of avelumab, an equal volume of 0.9% sodium chloride injection must be removed from the infusion bag and must be discarded.

13.1.4 Stability and Storage

Avelumab drug product must be stored at 2°C to 8°C until use, with a temperature log maintained daily. All medication boxes supplied to each study center must be stored carefully, safely, and separately from other drugs.

Avelumab drug product stored at room temperature (15°C to 25°C) or at elevated temperatures (38°C to 42°C) for extended periods is subject to degradation. Avelumab must not be frozen. Rough shaking of avelumab must be avoided.

Chemical and physical in-use stability has been demonstrated for 24 h at 25°C for the diluted medication in the approved infusion bags. However, from a microbiological point of view, the diluted solution should be used immediately and is not intended to be stored unless dilution has taken place in controlled and validated aseptic conditions.

13.1.5 Administration procedures

Please see section **3.4.2.1.**

13.1.6 Incompatibilities

The following treatments must not be administered during the study:

- Immunotherapy, immunosuppressive drugs (i.e., chemotherapy or systemic corticosteroids except for short term treatment of allergic reactions, for the treatment of irAEs, or for management of radiation induced tumor swelling), or other experimental pharmaceutical products. Short term administration of systemic steroid (i.e., for allergic reactions, the management of irAEs, and management of radiation induced tumor swelling) is allowed. Steroids with no or minimal systemic effect (topical, inhalation) are allowed.
- Any vaccine therapies for the prevention of infectious disease (e.g., seasonal flu vaccine, human papilloma virus vaccine) except administration of the inactive influenza vaccine.

13.2 AD-CEA VACCINE (BB IND# 17056)

13.2.1 Source

Ad-CEA will be supplied by the manufacturer, Etubics Corporation, through a Cooperative Research and Development Agreement (CRADA). The manufacturing department of Etubics will supply the vaccine, which will be distributed to the sites by Etubics Corporation.

13.2.2 Toxicity

A Phase I/II clinical trial of ETBX-011 (Ad5 [E1-, E2b-]-CEA(6D)) (IND#14325) that expresses the tumor-associated antigen carcinoembryonic antigen (CEA) has been performed. A summary of the study results on the clinical trial is presented below.

Schedule, Dosing, and Safety: The primary objective of the Phase I/II dosing trial was to assess safety and a secondary objective was to evaluate CEA-specific immune responses to CEA and to obtain preliminary data on response rate. The study was performed under an FDA-approved IND (IND14325) and registered at ClinicalTrials.gov (NCT01147965). Participants were recruited from oncology clinics at Duke University Medical Center and Medical Oncology Associates,

Spokane, WA and provided informed consent that was approved by IRB's. Patients with a histological confirmed diagnosis of metastatic malignancy who were previously treated with standard therapy known to have a possible survival benefit were enrolled into the study. For this study, the carcinoma must have had over expression of CEA as defined by immunohistochemical staining (at least 50% of the tumor with at least moderate intensity of staining) or a carcinoma known to be universally CEA positive *i.e.* colorectal adenocarcinoma). Patients were not treated until 4 or more weeks after any prior chemotherapy or radiation therapy. They could not have a history of autoimmune disease, serious intercurrent chronic or acute illness, active hepatitis, serologic evidence for HIV infection, or be receiving steroid or immunosuppressive therapy. All patients were ≥21 years old and had a Karnofsky Performance Score of 70% or higher and a life expectancy of at least 3 months. Pregnant women and nursing mothers were excluded. Dose Limiting Toxicities (DLTs) were defined as any Grade 3 or 4 immediate hypersensitivity reactions, Grade 3 or 4 fever that may possibly be associated with the immunization, Grade ≥2 autoimmune events except for vitiligo or fever for less than 2 days and less than 101.5 ºF, Grade ≥2 allergic reactions (Grade 2 is defined as generalized urticaria as defined by NCI Common Terminology Criteria for Adverse Events (CTCAE version 5.0), or Grade ≥3 non-hematologic toxicity. The Ad5 [E1-, E2b-]-CEA(6D) injections were given subcutaneously in the same thigh. The doses were administered every 3 weeks for 3 treatments as follows: Cohort 1 (3 patients) 10^9 VP in 0.5 ml; Cohort 2 (3 patients): 10^{10} VP; Cohort 3 (6 patients) 10^{11} VP. Following establishment of safety in Phase I, 12 additional patients were entered into a Phase II using the 10^{11} VP/dose. To evaluate a higher dose, 6 additional patients (cohort 5) received $5x10^{11}$ VP/dose.

Patient Demographics: One patient with CEA expressing bladder and one patient with lung carcinoma was enrolled. Thirty two patients, median age 57.5 (range 38-77) with metastatic colorectal cancer, who had failed a median of three prior chemotherapeutic regimens (range: 2–5), had a median performance status of 90% (range 70-100%), and had a median of three sites of metastatic disease (range 1-5), were enrolled. The majority was able to receive all three immunizations. Five patients who stopped immunizations early did so due to significant disease progression. The colorectal adenocarcinoma patient demographics compares favorably with previously published studies of patients with chemotherapy-refractory colorectal cancer [44-46].

Adverse Effects: A total of 94 immunization treatments were administered to all patients. There was no dose limiting toxicity and no serious adverse events that resulted in treatment discontinuation at any dose level. The most common toxicity was a self-limited, injection site reaction. Other reactions that occurred at a low frequency include fever, flu-like symptoms, anorexia, chills, nausea, and headache. These symptoms were also self-limiting and did not require intervention other than symptomatic measures such as acetaminophen. There were no SAE associated with immunizations. A summary of the adverse events reported on 34 patients treated and evaluated for safety are presented below.

Adverse Events: ≥ 5% Frequency Based on Incidence of Total Treatments

Adverse Events as of 07/31/12	# of Events	Unrelated/Unlikely	Possible	Probably/ Definite	Incidence % (based on 94 total treatments)

Injection Site Reaction	21	2		21	22.3
Flu-like symptoms	10	4	5	3	10.6
Fever	9	6	1	4	9.5
Fatigue	8	6	2		8.5
Shortness of Breath	6	6			6.3
Pain	6	4			6.3
Anorexia	5		1		5.3
Chills	5	5	1	4	5.3
Constipation	5	5			5.3
Edema	5	4			5.3
Nausea	5	4	1		5.3

Grade 3 and Grade 4 Adverse Events in ≥ 2% of Patients*

Adverse Events	Number of Grade 3 (G3)	Number of Grade 4 (G4)	Percent (Based on 34 patients evaluated for safety)
Pain (all types)	2		5.6
Fatigue	1		2.9
Anemia	1		2.9
Pleural effusion	1		2.9
Alkaline Phosphatase Increase	1		2.9
Abdominal Bloating	1		2.9
Bowel obstruction	1		2.9
GI disorder	1		2.9
Acute renal failure	1		2.9

*Represents reported adverse events whether or not related to ETBX-011 vaccine

Of 34 total patients entered into the trial, 28 received all three treatments and the blood hematology, chemistry, and ANA values at week 0 (prior to first treatment) were compared with those obtained at week 9 (three weeks after the third treatment). There were no biologically significant changes in chemistry, hematology, or ANA values.

13.2.3 Formulation and Preparation

ETBX-011 is a clear colorless liquid filled in a 2-mL amber vial containing 1.0 mL of extractable vaccine. There are total of $5x10^{11}$ total virus particles in 1.0 mL of the product. Each vial is sealed with a rubber stopper and has a white flip off seal. End user of the product will need to flip the white plastic portion of the cap up/off with their thumb to expose the rubber stopper,

and then puncture the stopper with an injection needle to withdraw the liquid. The rubber stopper is secured to the vial with an aluminum crimped seal.

13.2.4 Stability and Storage

Individual vials (in the desired number) of ETBX-011 will be packaged in a cardboard box and will be shipped over dry ice (<-80 °C) by overnight courier with a temperature monitoring device included. Upon receipt, one will inspect contents of package for any noticeable damages or defects. Unpack the shipment contents and place the cardboard box containing ETBX-011 vials into a freezer with a temperature control of ≤-20 °C. Receiver must stop the temperature monitoring device by turning off the power switch (instructions for handling and operation of temperature monitoring device will be provided with the package).

13.2.5 Administration Procedures

Please see section **3.4.2.2**

13.2.6 Incompatibilities

ETBX-011 is administered as a separate subcutaneous injection, with no known incompatibilities.

13.3 BEVACIZUMAB (AVASTIN)

(Please see package insert for complete information)

13.3.1 Source

Bevacizumab will be supplied by the local pharmacy using supply purchased from commercial sources.

13.3.2 Toxicity

The most common adverse reactions observed in bevacizumab patients at a rate > 10% and at least twice the control arm rate, are epistaxis, headache, hypertension, rhinitis, proteinuria, taste alteration, dry skin, rectal hemorrhage, lacrimation disorder, back pain and exfoliative dermatitis Some of the adverse reactions are commonly seen with chemotherapy; however, bevacizumab may exacerbate these reactions when combined with chemotherapeutic agents. Examples include palmar-plantar erythrodysaesthesia syndrome with pegylated liposomal doxorubicin or capecitabine peripheral sensory neuropathy with paclitaxel or oxaliplatin, and nail disorders or alopecia with paclitaxel.

13.3.2.1 Gastrointestinal Perforations and Fistulae

Serious and sometimes fatal gastrointestinal perforation occurs at a higher incidence in bevacizumab treated patients compared to controls. The incidence of gastrointestinal perforation ranged from 0.3 to 3.2% across clinical studies. From a clinical trial in patients with persistent, recurrent, or metastatic cervical cancer (Study 9), gastrointestinal perforations were reported in 3.2% of bevacizumab treated patients, all of whom had a history of prior pelvic radiation. Fatal outcome was reported in <1% of bevacizumab-treated patients. In a platinum-resistant ovarian cancer trial (Study 10), the incidence of GI perforation was 1.7% (3/179). In this trial, patients with evidence of recto-sigmoid involvement by pelvic examination or bowel involvement on CT scan or clinical symptoms of bowel obstruction were excluded.

13.3.2.2 Non-Gastrointestinal Fistulae

Serious and sometimes fatal fistula formation involving tracheo-esophageal, bronchopleural, biliary, vaginal, renal and bladder sites occurs at a higher incidence in bevacizumab -treated patients compared to controls. Uncommon (<1%) reports of fistulae that involve areas of the body other than the gastrointestinal tract were observed in clinical trials across various indications and have also been reported in post-marketing experience. Most events occurred within the first 6 months of bevacizumab therapy.

13.3.2.3 Surgery and Wound Healing Complications

Bevacizumab impairs wound healing in animal models. In clinical trials, administration of bevacizumab was not allowed until at least 28 days after surgery. In a controlled clinical trial, the incidence of wound healing complications, including serious and fatal complications, in patients with mCRC who underwent surgery during the course of bevacizumab treatment was 15% and in patients who did not receive bevacizumab, was 4%.

13.3.2.4 Hemorrhage

Bevacizumab can result in two distinct patterns of bleeding: minor hemorrhage, most commonly Grade 1 epistaxis; and serious, and in some cases fatal, hemorrhagic events. Severe or fatal hemorrhage, including hemoptysis, gastrointestinal bleeding, hematemesis, CNS hemorrhage, epistaxis, and vaginal bleeding occurred up to five-fold more frequently in patients receiving bevacizumab compared to patients receiving only chemotherapy. Across indications, the incidence of Grade ≥ 3 hemorrhagic events among patients receiving bevacizumab ranged from 0.4 to 6.9 %.

13.3.2.5 Arterial Thromboembolic Events

Serious, sometimes fatal, arterial thromboembolic events (ATE) including cerebral infarction, transient ischemic attacks, myocardial infarction, angina, and a variety of other ATE occurred at a higher incidence in patients receiving bevacizumab compared to those in the control arm. Across indications, the incidence of Grade ≥ 3 ATE in the bevacizumab containing arms was 2.6% compared to 0.8% in the control arms. Among patients receiving bevacizumab in combination with chemotherapy, the risk of developing ATE during therapy was increased in patients with a history of arterial thromboembolism, diabetes, or age greater than 65 years.

13.3.2.6 Hypertension

The incidence of severe hypertension is increased in patients receiving bevacizumab as compared to controls. Across clinical studies the incidence of Grade 3 or 4 hypertension ranged from 5-18%.

13.3.2.7 Posterior Reversible Encephalopathy Syndrome (PRES)

PRES has been reported with an incidence of < 0.5% in clinical studies. The onset of symptoms occurred from 16 hours to 1 year after initiation of bevacizumab. PRES is a neurological disorder which can present with headache, seizure, lethargy, confusion, blindness and other visual and neurologic disturbances. Mild to severe hypertension may be present. Magnetic resonance imaging (MRI) is necessary to confirm the diagnosis of PRES.

13.3.2.8 Proteinuria

The incidence and severity of proteinuria is increased in patients receiving bevacizumab as compared to controls. Nephrotic syndrome occurred in < 1% of patients receiving bevacizumab

in clinical trials, in some instances with fatal outcome. In a published case series, kidney biopsy of six patients with proteinuria showed findings consistent with thrombotic microangiopathy.

13.3.2.9 Infusion Reactions

Infusion reactions reported in the clinical trials and post-marketing experience include hypertension, hypertensive crises associated with neurologic signs and symptoms, wheezing, oxygen desaturation, Grade 3 hypersensitivity, chest pain, headaches, rigors, and diaphoresis. In clinical studies, infusion reactions with the first dose of bevacizumab were uncommon (< 3%) and severe reactions occurred in 0.2% of patients.

13.3.2.10 Ovarian Failure

The incidence of ovarian failure was higher (34% vs. 2%) in premenopausal women receiving bevacizumab in combination with mFOLFOX chemotherapy as compared to those receiving mFOLFOX chemotherapy alone for adjuvant treatment for colorectal cancer, a use for which bevacizumab is not approved. Inform females of reproductive potential of the risk of ovarian failure prior to starting treatment with bevacizumab

13.3.3 Formulation and Preparation

Bevacizumab is supplied as a clear to slightly opalescent, sterile liquid ready for parenteral administration in vials of two sizes:

- 100 mg/4 mL, single use vial
- 400 mg/16 mL, single use vial

Use appropriate aseptic technique. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration, whenever solution and container permit. Withdraw necessary amount of bevacizumab and dilute in a total volume of 100 mL of 0.9% Sodium Chloride Injection, USP. Discard any unused portion left in a vial, as the product contains no preservatives.

Do not administer or mix with dextrose solution.

13.3.4 Stability and Storage

Bevacizumab vials should be stored according to FDA-approved labeling. Do not freeze or shake.

Diluted bevacizumab solutions may be stored at 2−8°C (36−46°F) for up to 24 hours, per site pharmacy guidelines. Store in the original carton until time of use. No incompatibilities between bevacizumab and polyvinylchloride or polyolefin bags have been observed.

13.3.5 Administration Procedures

Please see section **3.4.1.**

13.3.6 Incompatibilities

A drug interaction study was performed in which irinotecan was administered as part of the FOLFIRI regimen with or without bevacizumab. The results demonstrated no significant effect of bevacizumab on the pharmacokinetics of irinotecan or its active metabolite SN38.

In a study assessing the safety and efficacy of bevacizumab as first line therapy in non-small cell lung cancer, based on limited data, there did not appear to be a difference in the mean exposure of either carboplatin or paclitaxel when each was administered alone or in combination with bevacizumab. However, 3 of the 8 patients receiving bevacizumab plus paclitaxel/carboplatin had substantially lower paclitaxel exposure after four cycles of treatment (at Day 63) than those at Day 0, while patients receiving paclitaxel/carboplatin without bevacizumab had a greater paclitaxel exposure at Day 63 than at Day 0.

13.4 CAPECITABINE

(Please see package insert for complete information)

13.4.1 Source

Capecitabine will be supplied by the local pharmacy using supply purchased from commercial sources.

13.4.2 Toxicity

Most common adverse reactions (≥30%) were diarrhea, hand-and-foot syndrome, nausea, vomiting, abdominal pain, fatigue/weakness, and hyperbilirubinemia. Other adverse reactions, including serious adverse reactions, have been reported.

Pooled Phase 3 Colorectal Trials: Percent Incidence of Adverse Reactions in ≥5% of Patients

Adverse Event	Capecitabine (n=596)		
	Total %	Grade 3 %	Grade 4 %
Number of Patients With > One Adverse Event	96	52	9
Body System/Adverse Event			
GI			
Diarrhea	55	13	2
Nausea	43	4	–
Vomiting	27	4	<1
Stomatitis	25	2	<1
Abdominal Pain	35	9	<1
Gastrointestinal Motility Disorder	10	<1	–
Constipation	14	1	<1
Oral Discomfort	10	–	–
Upper GI Inflammatory Disorders	8	<1	–
Gastrointestinal Hemorrhage	6	1	<1
Ileus	6	4	1
Skin and Subcutaneous			
Hand-and-Foot Syndrome	54	17	NA
Dermatitis	27	1	–

Adverse Event	**Capecitabine (n=596)**		
	Total %	**Grade 3 %**	**Grade 4 %**
Number of Patients With > One Adverse Event	96	52	9
Body System/Adverse Event			
Skin Discoloration	7	<1	–
Alopecia	6	–	–
General			
Fatigue/Weakness	42	4	–
Pyrexia	18	1	–
Edema	15	1	–
Pain	12	1	–
Chest Pain	6	1	–
Neurological			
Peripheral Sensory Neuropathy	10	–	–
Headache	10	1	–
Dizziness*	8	<1	–
Insomnia	7	–	–
Taste Disturbance	6	1	–
Metabolism			
Appetite Decreased	26	3	<1
Dehydration	7	2	<1
Eye			
Eye Irritation	13	–	–
Vision Abnormal	5	–	–
Respiratory			
Dyspnea	14	1	–
Cough	7	<1	1
Pharyngeal Disorder	5	–	–
Epistaxis	3	<1	–
Sore Throat	2	–	–
Musculoskeletal			
Back Pain	10	2	–
Arthralgia	8	1	–

Adverse Event	Capecitabine (n=596)		
	Total %	Grade 3 %	Grade 4 %
Number of Patients With > One Adverse Event	96	52	9
Body System/Adverse Event			
Vascular			
Venous Thrombosis	8	3	<1
Psychiatric			
Mood Alteration	5	–	–
Depression	5	–	–
Infections			
Viral	5	<1	–
Blood and Lymphatic			
Anemia	80	2	<1
Neutropenia	13	1	2
Hepatobiliary			
Hyperbilirubinemia	48	18	5

– Not observed
* Excluding vertigo

NA = Not Applicable

13.4.3 Formulation and preparation

Tablets: 150 mg and 500 mg

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.

13.4.4 Stability and Storage

Store at 25°C (77°F); excursions permitted to 15° to 30°C (59° to 86°F). KEEP TIGHTLY CLOSED.

Care should be exercised in the handling of capecitabine. Capecitabine tablets should not be cut or crushed. Procedures for the proper handling and disposal of anticancer drugs should be considered. Any unused product should be disposed of in accordance with local requirements, or drug take back programs. Several guidelines on the subject have been published.

13.4.5 Administration procedures

Please see section **3.4.1.2**

13.4.6 Incompatibilities

- Anticoagulants: Monitor anticoagulant response (INR or prothrombin time) frequently in order to adjust the anticoagulant dose as needed.
- Phenytoin: Monitor phenytoin levels in patients taking capecitabine concomitantly with phenytoin. The phenytoin dose may need to be reduced.
- Leucovorin: The concentration of 5-fluorouracil is increased and its toxicity may be enhanced by leucovorin.
- CYP2C9 substrates: Care should be exercised when capecitabine is coadministered with CYP2C9 substrates.
- Food reduced both the rate and extent of absorption of capecitabine.

13.5 5-FLUOROURACIL (5-FU)

(Please see package insert for complete information)

13.5.1 Source

5-FU will be supplied by the local pharmacy using supply purchased from commercial sources.

13.5.2 Toxicity

5-FU should be used with extreme caution in poor risk patients with a history of high-dose pelvic irradiation or previous use of alkylating agents, those who have a widespread involvement of bone marrow by metastatic tumors or those with impaired hepatic or renal function.

Rarely, unexpected, severe toxicity (e.g., stomatitis, diarrhea, neutropenia and neurotoxicity) associated with 5-fluorouracil has been attributed to deficiency of dipyrimidine dehydrogenase activity. A few patients have been rechallenged with 5-fluorouracil and despite 5-fluorouracil dose lowering, toxicity recurred and progressed with worse morbidity. Absence of this catabolic enzyme appears to result in prolonged clearance of 5-fluorouracil.

The administration of 5-fluorouracil has been associated with the occurrence of palmar-plantar erythrodysesthesia syndrome, also known as hand-foot syndrome. Interruption of therapy is followed by gradual resolution over 5 to 7 days. Although pyridoxine has been reported to ameliorate the palmar-plantar erythrodysesthesia syndrome, its safety and effectiveness has not been established.

Stomatitis and esophagopharyngitis (which may lead to sloughing and ulceration), diarrhea, anorexia, nausea and emesis are commonly seen during therapy.

Leukopenia usually follows every course of adequate therapy with fluorouracil. The lowest white blood cell counts are commonly observed between the 9th and 14th days after the first course of treatment, although uncommonly the maximal depression may be delayed for as long as 20 days. By the 30th day the count has usually returned to the normal range.

Alopecia and dermatitis may be seen in a substantial number of cases. The dermatitis most often seen is a pruritic maculopapular rash usually appearing on the extremities and less frequently on the trunk. It is generally reversible and usually responsive to symptomatic treatment.

Other adverse reactions are:

- Hematologic: pancytopenia, thrombocytopenia, agranulocytosis, anemia.
- Cardiovascular: myocardial ischemia, angina.
- Gastrointestinal: gastrointestinal ulceration and bleeding.
- Allergic Reactions: anaphylaxis and generalized allergic reactions.
- Neurologic: acute cerebellar syndrome (which may persist following discontinuance of treatment), nystagmus, headache.
- Dermatologic: dry skin; fissuring; photosensitivity, as manifested by erythema or increased pigmentation of the skin; vein pigmentation; palmar-planter erythrodysesthesia syndrome, as manifested by tingling of the hands and feet followed by pain, erythema and swelling.
- Ophthalmic: lacrimal duct stenosis, visual changes, lacrimation, photophobia.
- Psychiatric: disorientation, confusion, euphoria.
- Miscellaneous: thrombophlebitis, epistaxis, nail changes (including loss of nails)

13.5.3 Formulation and preparation

13.5.3.1 Formulation

Fluorouracil injection, USP an antineoplastic antimetabolite, is a sterile, nonpyrogenic injectable solution for intravenous administration. Each 10 mL contains 500 mg fluorouracil; pH is adjusted to approximately 9.2 with sodium hydroxide.

For intravenous use. Fluorouracil injection, USP is available at a concentration of 50mg/mL.

The 50 mL and 100 mL pharmacy bulk packages are packaged 5 vials per shelf pack.

13.5.3.2 Preparation

No dilution is required.

13.5.4 Stability and Storage

Store at room temperature 15° to 30°C (59° to 86°F). Protect from light. Retain in carton until time of use.

13.5.5 Administration procedures

Please see section **3.4.1.1.**

13.5.6 Incompatibilities

Leucovorin calcium may enhance the toxicity of fluorouracil.

13.6 LEUCOVORIN

(Please see package insert for complete information)

13.6.1 Source

Leucovorin will be supplied by the local pharmacy using supply purchased from commercial sources. In the event of a leucovorin drug shortage, levoleucovorin may be substituted using equivalent doses at the investigator's discretion and administered according to FDA-approved labeling.

13.6.2 Toxicity

Allergic sensitization, including anaphylactoid reactions and urticaria, has been reported following the administration of both oral and parenteral leucovorin. No other adverse reactions have been attributed to the use of leucovorin per se.

Table 1 below summarizes significant adverse events occurring in 316 patients treated with the leucovorin/5-fluorouracil combinations compared against 70 patients treated with 5-fl uorouracil alone for advanced colorectal carcinoma. These data are taken from the Mayo/NCCTG large multicenter prospective trial evaluating the efficacy and safety of the combination regimen.

Table 1 : Percentage of Patients Treated with Leucovorin/Fluorouracil for Advanced Colorectal Carcinoma Reporting Adverse Experiences or Hospitalized for Toxicity

Toxicity	**(High LV) /5-FU (N=155)**		**(Low LV) /5-FU (N=161)**		**5-FU Alone (N=70)**	
	Any (%)	**Grade 3+ (%)**	**Any (%)**	**Grade 3+ (%)**	**Any (%)**	**Grade 3+ (%)**
Leukopenia	69	14	83	23	93	48
Thrombocytopenia	8	2	8	1	18	3
Infection	8	1	3	1	7	2
Nausea	74	10	80	9	60	6
Vomiting	46	8	44	9	40	7
Diarrhea	66	18	67	14	43	11
Stomatitis	75	27	84	29	59	16
Constipation	3	0	4	0	1	-
Lethargy/Malaise/Fatigue	13	3	12	2	6	3
Alopecia	42	5	43	6	37	7
Dermatitis	21	2	25	1	13	-
Anorexia	14	1	22	4	14	-
Hospitalization for Toxicity	5%		15%		7%	

Toxicity	(High LV) /5-FU (N=155)		(Low LV) /5-FU (N=161)		5-FU Alone (N=70)	
	Any (%)	Grade 3+ (%)	Any (%)	Grade 3+ (%)	Any (%)	Grade 3+ (%)
High LV = Leucovorin 200 mg/m2, Low LV = Leucovorin 20 mg/m2 Any = percentage of patients reporting toxicity of any severity Grade 3+ = percentage of patients reporting toxicity of Grade 3 or higher						

13.6.3 Formulation and preparation

13.6.3.1 Formulation

Leucovorin Calcium Injection USP is a sterile, preservative-free solution indicated for intramuscular (IM) or intravenous (IV) administration in a 50 mL single-dose vial. Each mL contains leucovorin calcium equivalent to 10 mg Leucovorin, USP; 8 mg sodium chloride; sodium hydroxide and/or hydrochloric acid for pH adjustment pH 7.8 (6.5 to 8.5).

There is 0.004 mEq of calcium per mg of leucovorin. Solution contains no bacteriostat or antimicrobial agents.

Leucovorin Calcium for Injection is a sterile product indicated for intramuscular (IM) or intravenous (IV) administration and is supplied in 50 mg, 100 mg, 200 mg, and 350 mg vials.

Each 50 mg vial of Leucovorin Calcium for Injection, when reconstituted with 5 mL of sterile diluent, contains leucovorin (as the calcium salt) 10 mg/mL.

Each 100 mg vial of Leucovorin Calcium for Injection, when reconstituted with 10 mL of sterile diluent, contains leucovorin (as the calcium salt) 10 mg/mL.

Each 200 mg vial of Leucovorin Calcium for Injection, when reconstituted with 20 mL of sterile diluent, contains leucovorin (as the calcium salt) 10 mg/mL.

Each 350 mg vial of Leucovorin Calcium for Injection, when reconstituted with 17.5 mL of sterile diluent, contains leucovorin (as the calcium salt) 20 mg/mL.

In each dosage form, one milligram of leucovorin calcium contains 0.002 mmol of leucovorin and 0.002 mmol of calcium.

13.6.3.2 Preparation

These lyophilized products contain no preservative. The inactive ingredient is Sodium Chloride, USP, added to adjust tonicity.

Because of the benzyl alcohol contained in certain diluents used for reconstituting Leucovorin Calcium for Injection, when doses greater than 10 mg/m2 (as in this study) are administered, Leucovorin Calcium for Injection should be reconstituted with Sterile Water for Injection, USP, and used immediately.

13.6.4 Stability and Storage

Store at 20°C to 25°C (68° to 77° F). [See USP Controlled Room Temperature.] **Protect from light.** Retain in carton until time of use.

Reconstituted form should be used immediately.

13.6.5 Administration procedures

Please see section **3.4.1.1.**

13.6.6 Incompatibilities

Folic acid in large amounts may counteract the antiepileptic effect of phenobarbital, phenytoin and primidone, and increase the frequency of seizures in susceptible pediatric patients.

Preliminary animal and human studies have shown that small quantities of systemically administered leucovorin enter the CSF primarily as 5-methyltetrahydrofolate and, in humans, remain 1 to 3 orders of magnitude lower than the usual methotrexate concentrations following intrathecal administration. However, high doses of leucovorin may reduce the efficacy of intrathecally administered methotrexate.

Leucovorin may enhance the toxicity of 5-fluorouracil

13.7 OXALIPLATIN

(Please see package insert for complete information)

13.7.1 Source

Oxaliplatin will be supplied by the local pharmacy using supply purchased from commercial sources.

13.7.2 Toxicity

The most common side effects of oxaliplatin include:

- neutropenia, anemia, thrombocytopenia, hypertension, diarrhea, nausea, vomiting, constipation, oral mucositis, abdominal pain, anorexia, fatigue, injection site reactions, alopecia and dehydration

Potentially serious toxicities include:

- **Grade 3/4 hypersensitivity,** including anaphylactic/anaphylactoid reactions, to oxaliplatin has been observed in 2-3% of colon cancer patients. These reactions are usually managed with standard epinephrine, corticosteroid, antihistamine therapy, and require discontinuation of therapy or desensitization protocol for continuation of therapy.
- **Peripheral sensory neuropathy** (acute ≤ 14 days and persistent > 14 days) was reported in adjuvant patients treated with ELOXATIN as part of FOLFOX with a frequency of 92% (all grades) and 13% (grade 3). At the 28-day follow-up after the last treatment cycle, 60% of all patients had any grade (Grade 1=40%, Grade 2=16%, Grade 3=5%) peripheral sensory neuropathy decreasing to 39% at 6 months follow-up (Grade 1=31%, Grade 2=7%, Grade 3=1%) and 21% at 18 months of follow-up (Grade 1=17%, Grade 2=3%, Grade 3=1%).

Overall, neuropathy was reported in patients previously untreated for advanced colorectal cancer in 82% (all grades) and 19% (grade 3/4), and in the previously treated patients in 74% (all grades) and 7% (grade 3/4) events. Information regarding reversibility of neuropathy was not available from the trial for patients who had not been previously treated for colorectal cancer.

- **Reversible Posterior Leukoencephalopathy Syndrome** (RPLS, also known as PRES, Posterior Reversible Encephalopathy Syndrome) has been observed in clinical trials (< 0.1%) and postmarketing experience. Signs and symptoms of RPLS could be headache, altered mental functioning, seizures, abnormal vision from blurriness to blindness, associated or not with hypertension. Diagnosis of RPLS is based upon confirmation by brain imaging.
- **Pulmonary Toxicity.** oxaliplatin has been associated with pulmonary fibrosis (<1% of study patients), which may be fatal. The combined incidence of cough and dyspnea was 7.4% (any grade) and < 1% (grade 3) with no grade 4 events in the oxaliplatin plus infusional 5-fluorouracil/leucovorin arm compared to 4.5% (any grade) and no grade 3 and 0.1% grade 4 events in the infusional 5- fluorouracil/leucovorin alone arm in adjuvant colon cancer patients. In this study, one patient died from eosinophilic pneumonia in the oxaliplatin combination arm. The combined incidence of cough, dyspnea and hypoxia was 43% (any grade) and 7% (grade 3 and 4) in the oxaliplatin plus 5-fluorouracil/leucovorin arm compared to 32% (any grade) and 5% (grade 3 and 4) in the irinotecan plus 5-fluorouracil/leucovorin arm of unknown duration for patients with previously untreated colorectal cancer. In case of unexplained respiratory symptoms such as non-productive cough, dyspnea, crackles, or radiological pulmonary infiltrates, ELOXATIN should be discontinued until further pulmonary investigation excludes interstitial lung disease or pulmonary fibrosis.
- **Hepatotoxicity** as evidenced in the adjuvant study, by increase in transaminases (57% vs. 34%) and alkaline phosphatase (42% vs. 20%) was observed more commonly in the oxaliplatin combination arm than in the control arm. The incidence of increased bilirubin was similar on both arms. Changes noted on liver biopsies include: peliosis, nodular regenerative hyperplasia or sinusoidal alterations, perisinusoidal fibrosis, and veno-occlusive lesions. Hepatic vascular disorders should be considered, and if appropriate, should be investigated in case of abnormal liver function test results or portal hypertension, which cannot be explained by liver metastases

13.7.3 Formulation and preparation

13.7.3.1 Formulation

13.7.3.1.1 Powder

Oxaliplatin is supplied in single-use vials containing 50 mg or 100 mg of oxaliplatin as a sterile, preservative-free lyophilized powder for reconstitution. Lactose monohydrate is also present as an inactive ingredient.

13.7.3.1.2 Solution

Oxaliplatin is supplied in single-use vials containing 50 mg, 100 mg or 200 mg of oxaliplatin as a sterile, preservative-free, aqueous solution at a concentration of 5 mg/ml. Water for Injection, USP is present as an inactive ingredient.

13.7.3.2 Preparation

13.7.3.2.1 Powder

Reconstitution or final dilution must never be performed with a sodium chloride solution or other chloride containing solutions.

The lyophilized powder is reconstituted by adding 10 mL (for the 50 mg vial) or 20 mL (for the 100 mg vial) of Water for Injection, USP or 5% Dextrose Injection, USP. Do not administer the reconstituted solution without further dilution. The reconstituted solution must be further diluted in an infusion solution of 250-500 mL of 5% Dextrose Injection, USP.

After reconstitution in the original vial, the solution may be stored up to 24 hours under refrigeration [2-8°C (36-46°F)]. After final dilution with 250-500 mL of 5% Dextrose Injection, USP, the shelf life is 6 hours at room temperature [20-25°C (68-77°F)] or up to 24 hours under refrigeration [2-8°C (36-46°F)].

Oxaliplatin is not light sensitive.

13.7.3.2.2 Solution

Do not freeze and protect from light the concentrated solution.

A final dilution must never be performed with a sodium chloride solution or other chloride containing solutions.

The solution must be further diluted in an infusion solution of 250-500 mL of 5% Dextrose Injection, USP.

After dilution with 250-500 mL of 5% Dextrose Injection, USP, the shelf life is 6 hours at room temperature [20-25°C (68-77°F)] or up to 24 hours under refrigeration [2-8°C (36-46°F)].

After final dilution, protection from light is not required.

Oxaliplatin is incompatible in solution with alkaline medications or media (such as basic solutions of 5-fluorouracil) and must not be mixed with these or administered simultaneously through the same infusion line. The infusion line should be flushed with 5% Dextrose Injection, USP prior to administration of any concomitant medication.

13.7.3.2.3 General

Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration and discarded if present.

Needles or intravenous administration sets containing aluminum parts that may come in contact with oxaliplatin should not be used for the preparation or mixing of the drug. Aluminum has been reported to cause degradation of platinum compounds.

13.7.4 Stability and Storage

13.7.4.1 Powder for solution for infusion:

Store under normal lighting conditions at 25°C (77°F); excursions permitted to 15-30°C (59-86°F).

13.7.4.2 Concentrate for solution for infusion:

Store at 25°C (77°F); excursions permitted to 15-30°C (59-86°F). Do not freeze and protect from light (keep in original outer carton).

13.7.5 Administration procedures

Please see section **3.4.1.1.**

13.7.6 Incompatibilities

No specific cytochrome P-450-based drug interaction studies have been conducted. No pharmacokinetic interaction between 85 mg/m2 oxaliplatin and 5-fluorouracil/leucovorin has been observed in patients treated every 2 weeks. Increases of 5-fluorouracil plasma concentrations by approximately 20% have been observed with doses of 130 mg/m2 oxaliplatin dosed every 3 weeks. Because platinum-containing species are eliminated primarily through the kidney, clearance of these products may be decreased by co-administration of potentially nephrotoxic compounds; although, this has not been specifically studied.

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15 APPENDICES

15.1 APPENDIX A: PRECLINICAL MODEL EVALUATION OF THE COMBINATION OF FOLFOX WITH ANTI-PD-L1

MSB0010718C Combination with FOLFOLX in the MC38 Tumor Model
Study ID: IONC20042011CKH (TI10-079, TI11-001)

Pre-clinical Pharmacology – TA ImmunoOncology

Non-clinical STUDY REPORT, *In Vivo*

Combination of MSB0010718C (Anti-PD-L1) and FOLFOLX in a Subcutaneous MC38 Tumor Model in C57BL/6 Mice

Project name:	Anti-PD-L1
Project code:	G.10007001
Study number:	IONC20012011CKH (TI10-079, TI11-001)
Authors:	Huakui Yu, Robert Tighe, Ken Hance
GLP Compliance :	The study was not conducted in compliance with Good Laboratory Practice (GLP).
Test facility:	Yan Lan, Director Translational Immunology TA-Oncology-Immunotherapy EMD Serono Research Institute 45A Middlesex Turnpike Billerica, MA 01821
Date of report creation:	April 20th, 2011

15.2 APPENDIX B-PERFORMANCE STATUS CRITERIA

ECOG Performance Status Scale		**Karnofsky Performance Scale**	
Grade	Descriptions	Percent	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.	100	Normal, no complaints, no evidence of disease.
		90	Able to carry on normal activity; minor signs or symptoms of disease.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (e.g., light housework, office work).	80	Normal activity with effort; some signs or symptoms of disease.
		70	Cares for self, unable to carry on normal activity or to do active work.
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.	60	Requires occasional assistance, but is able to care for most of his/her needs.
		50	Requires considerable assistance and frequent medical care.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	40	Disabled, requires special care and assistance.
		30	Severely disabled, hospitalization indicated. Death not imminent.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	20	Very sick, hospitalization indicated. Death not imminent.
		10	Moribund, fatal processes progressing rapidly.
5	Dead.	0	Dead.

15.3 APPENDIX C: CCR PROBLEM REPORT FORM

CCR REPORTABLE EVENT FORM (REF)

NCI Protocol #: Click or tap here to enter text.	
Protocol Title: Click or tap here to enter text.	
Report version: *(select one)* ☐ Initial Report ☐ Follow-up	
Site Principal Investigator: Click or tap here to enter text.	
Date site PI was notified of the problem: Click or tap to enter a date.	**Date of problem:** Click or tap to enter a date.
If delay in reporting to the coordinating center, please explain: Click or tap here to enter text.	
Location of problem: *(e.g., patient's home, doctor's office)* Click or tap here to enter text.	
Description of Subject Does this problem apply to a subject? ☐ yes ☐ not applicable (more than one subject is involved) **If yes, enter details below:** Subject ID: Click or tap here to enter text. (*do not use medical record number)* Sex: ☐ Male ☐ Female Age: Click or tap here to enter text.	

Diagnosis: Click or tap here to enter text.

Name the problem: *(select all that apply)*

☐ Specimen collection issue

☐ Informed consent issue

☐ Ineligible for enrollment

☐ Breach of PII

☐ Other, briefly state the nature of the problem: Click or tap here to enter text.

Detailed Description of the problem: (*Include any relevant treatment, outcomes or pertinent history*): Click or tap here to enter text.

What are you reporting?

☐ **unanticipated problem**

☐ **death**

☐ **non-compliance (other than a protocol deviation)**

☐ **protocol deviation**

☐ **new information that might affect the willingness of subjects to enroll or continue participation in this study**

If interventional or expanded access study, please answer the following questions about your site:

How many participants are still receiving the study intervention? Click or tap here to enter text. How many participants completed study interventions but remain in follow up? Click or tap here to enter text. How many participants are enrolled but not yet receiving study interventions? Click or tap here to enter text.
Have similar problems occurred on this protocol at your site? **☐ Yes ☐ No**
Describe what steps you have already taken or will be taking as a result of this problem: Click or tap here to enter text.

INVESTIGATOR'S SIGNATURE:	**DATE:**

