WWAMI
School of Medical Education
University of Alaska Anchorage

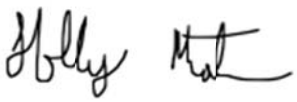
3211 Providence Drive, HSB 301
Anchorage, Alaska 99508-4614
T 907.786.4789 • F 907.786.4700
www.uaa.alaska.edu/wwami

Date: May 8, 2018

Dear Ze-Mao Gong and Reviewers,

On behalf of my fellow co-author's, I am resubmitting the revised manuscript #39147 entitled "**Gastric cancer in Alaska Native people: A cancer health disparity**". We would like to thank the Reviewers for their thoughtful critique of our submitted manuscript. Many good points were brought up by all four Reviewers. Below, we provide point by point response to the Reviewer's comments. If any additional information is required in order for your editorial board to make a decision regarding this manuscript, please do not hesitate to contact me.

Sincerely,



Holly Martinson, PhD
Assistant Professor
WWAMI School of Medical Education
University of Alaska Anchorage
Phone: 1-907-786-4672
hamartinson@alaska.edu

REVIEWER ONE COMMENTS TO AUTHORS

Reviewer One Major Comment: *Signet ring cell and poor survival may be attributable to that AN people may visit hospital at later stage of gastric cancer. But younger age and different location was not explained by this hypothesis. Was there any speculation to these results- younger age and location?*

Response to Reviewer comment younger age at diagnosis and location: We appreciate the Reviewer's comment on further speculating why we observed a younger age at time of diagnosis for Alaska Native (AN) gastric cancer patients. The Reviewer brings up two important etiological factors that distinguish gastric cancer as a unique cancer health disparity among the Alaska Native people. Firstly, based on extensive understanding of the promotional role of *Helicobacter pylori* in gastric cancer, it is widely known that exposure to *H. pylori* is primary responsible for the increased incidence, non-cardia location, and possibly earlier age of diagnosis. In a study performed by the Centers for Disease Control, they determined AN children were infected with *H. pylori* early in life, often by the age of 4. We have also included in the discussion other factors that could play a role in younger age at diagnosis such as early exposure to tobacco, genetic predisposition, and early age of diagnosis of other cancer types among Alaska Native people. In order to address this comment, we have added the following paragraph to the discussion:

> The younger age at diagnosis among AN patients with gastric cancer could be driven by multiple factors. One factor is earlier exposure to particular gastric cancer risk factors such as *H. pylori* infection and tobacco use. Previous research revealed 40% of AN children have been infected with H. pylori by age 4, 70% by age 10, and 78% by age 14 (Parkinson et al. 2000). This study and our results suggest the likelihood of long term exposure to systemic inflammation due to the early age of acquisition of *H. pylori* may play an important role in the high incidence of non-cardia cancer, younger age at diagnosis and the overall cancer health disparity among the AN people. Further, the high prevalence of tobacco

use among the AN people may also contribute to the younger age of diagnosis in gastric cancer patients. Another factor associated with a younger age of gastric cancer diagnosis is genetic predisposition such as CDH1 germline mutations that result in hereditary diffuse gastric cancers. Approximately 30% of AN patients had a family history of gastrointestinal cancers and there was no difference in age of diagnosis. Further, other types of cancer among the AN people such as lung, kidney, and colorectal cancer are also associated with younger age of diagnosis suggesting earlier age of diagnosis of cancer is a general characteristic in AN cancer patients compared to NHW patients. Often cancers diagnosed at a younger age are more aggressive and are found at a later stage, which may also contribute to cancer health disparities among the AN people.

Reviewer One Major Comment: *Were there any differences between Alaska and the other states in the US in terms of medical systems? Number of hospitals per people, medical insurance company, average distance of patients and hospitals.*

Response to Reviewer comment on medical system, number of hospitals, medical insurance, and patient distance from hospital: The Reviewer brings up an important point regarding how differences in patient care and access to a hospital may affect cancer rates among the Alaska Native people. The United States government provides health care to the Alaska Native and American Indian populations through the Indian Health Service (IHS). The IHS provides comprehensive health services to all Alaska Native people through the Alaska Native Tribal Health Consortium (ANTHC) whom oversees 99% of the Alaska Area IHS budget and serves all 228 federally recognized Alaska tribes. In Alaska, there are 58 tribal health centers, 160 tribal community health aide clinics, and one state-wide referral hospital the Alaska Native Medical Center (ANMC), that oversees specialty care such as, medical oncology. ANMC, located in Anchorage, AK, includes a 167-bed hospital that provides specialty medical care to 150,000 Alaska Native people. When patients are diagnosed with cancer they must fly (many communities are not on the road system) from their communities to receive their

cancer care at ANMC. For some patients traveling to and from their communities in order to receive their care and treatments may be a barrier even though their health care is provided by IHS. There has been little research on how the average distance of patients from the state-wide referral hospital (ANMC) affects cancer patient care. This is an important area of study and is worthy of further investigation. Below, we have addressed this comment by adding the following paragraph to the discussion:

> The Alaska Tribal Health System is a unique health system with 58 tribal health centers, 160 tribal community health aide clinics, and six regional hospitals dispersed throughout a vast land mass that covers more than 25% of the contiguous US. Patients with cancer are referred to the Alaska Native Medical Center, a tertiary hospital in Anchorage, Alaska where they receive cancer therapies according to standard international guidelines[18]. Many of the AN patients included in this study must travel to ANMC to receive their care and medical treatments. The average patient distance from ANMC and its effect on patient care and outcomes has not been studied but is worthy of further investigation.

Reviewer One Major Comment: *As stated in Discussion, number of study subjects were different. NHW=40717, AN=132. What specific affect to results did the authors have in mind regarding this point?*

Response to Reviewer comment number of study subjects: We understand the Reviewer's concerns that the number of AN people included in this study, 132 individuals, is small. In order to conduct this study, we identified all AN gastric cancer patients diagnosed from 2006-2014 at the Alaska Native Medical Center (ANMC). ANMC is the only state-wide referral hospital that oversees specialty care such as, medical oncology therefore all AN cancer patients receive their oncology care at ANMC

or elect to receive their care at a private hospital. To further address this comment, we have added the following sentence to the discussion:

> The AN population is relatively small- consisting of 150,000 people. In order to conduct this study, we reviewed all AN gastric cancer cases diagnosed at the Alaska Native Medical Center between 2006-2014. Approximately 132 patients were identified that had the epidemiological information needed to conduct this study. Even with the small number of cases, we were able to detect significant differences in the results. Although the AN population is small, we feel this population is worthy of study because of the poor clinical outcomes and gastric cancer mortality rates that are unique to this population within Alaska.

Reviewer One Major Comment: *Was there any screening system of gastric cancer in Alaska or the other states of the US? For example, upper gastrointestinal series.*

Response to Reviewers comment earlier diagnosis and screening system: Reviewer 1 commented on differences in gastric cancer screening in Alaska compared to the US and the world, raising a very important point. The screening programs implemented in Asian countries in areas of high disease rates have reduced the high mortality rates of gastric cancer among their people. However, the National Cancer Institute (NCI) PDQ cancer information summary for gastric cancer screening, no major US organization recommends general population screening for gastric cancer because there is no evidence that screening would results in a decrease in mortality in areas of low incidence of the disease such as the US. Among the total US population gastric cancer is the 15th most common cancer and represents 1.5% of all new cancer cases, in contrast to the AN population it's the 5th most common cancer and represents 5.6% of all new cancer cases. We are currently collaboration with the Centers for Disease Control to better understand which people are more likely to be diagnosed with gastric cancer and whether to implement standard guidelines on screening for high risk patients in order

to reduce mortality rates. For example, patients with a previous history of *H. pylori* infection or chronic gastritis may need to be closely monitored for reinfection or referred for an endoscopy. We addressed this comment by adding the following sentence to the discussion:

> There are currently no standard guidelines on screening for gastric cancer in the US[18], whereas Asian countries with a high incidence of gastric cancer have implemented screening programs using a variety of modalities. However, the most effective gastric cancer screening modality and the screening interval remains controversial.

Reviewer One Major Comment: *Patient survival was longer in those with chronic gastritis. Was there any speculation to this result? It is expected that early diagnosis would improve patient survival.*

Response to Reviewer comment chronic gastritis: We appreciate the Reviewers comments to further evaluate why chronic gastritis is associated with better overall survival in our patient population. We evaluated whether patients without chronic gastritis were more likely to be diagnosed at a later stage and discovered 75% of AN patients without gastritis were diagnosed at stage IV compared to 43% of AN patients with chronic gastritis.

The following sentence was added to the results:

> Upon further investigation, patients were more likely to be diagnosed with stage IV cancer without gastritis (75%) compared to patients with chronic gastritis (43%).

The following paragraph was added to the discussion:

AN patients with the presence of chronic gastritis were shown to have a more favorable prognosis, which was also associated with an earlier stage at diagnosis. This result suggests that AN patients presenting with symptoms of chronic gastritis may be at higher risk for developing gastric cancer and may benefit from an endoscopy at time of initial presentation.

REVIEWER TWO COMMENTS TO AUTHORS

Reviewer Two Major Comment: *However, it is very difficult to analyze the features of a cancer on a specific population evaluating only the clinical characteristics of 132 patients. Furthermore, representative well-designed studies are needed to give some conclusion.*

Response to Reviewer comment on number of study subjects: The Reviewer brings up an important point that was also addressed by Reviewer one's comments. The number of Alaska Native (AN) people included in this study, 132 individuals, is small. In order to conduct this study, we identified all AN gastric cancer patients diagnosed from 2006-2014 at the Alaska Native Medical Center (ANMC). ANMC is the only state-wide referral hospital that oversees specialty care such as, medical oncology therefore all AN cancer patients receive their oncology care at ANMC or elect to receive their care at a private hospital. To further address this comment, we have added the following sentence to the discussion:

The AN population is relatively small- consisting of 150,000 people. In order to conduct this study, we reviewed all AN gastric cancer cases diagnosed at the Alaska Native Medical Center (ANMC) between 2006-2014, approximately 132 cases. Records from this timespan had the epidemiological information needed to conduct this study, which is why we focused on these individuals. Even with the small number of cases, we were able to detect significant differences in the

results. Although the AN population is small, we feel this population is worthy of study because of the poor clinical outcomes and gastric cancer mortality rates that are unique to this population within Alaska.

REVIEWER THREE COMMENTS TO AUTHORS

Reviewer Three Major Comment: *There is definitely an issue with this retrospective, surveillance study especially in the year 2018 with an original title: "Gastric cancer in Alaska Native people: A cancer health disparity" considering the title referred and approved in the Institutional Review Board Approval Form : "Molecular Characterization of Gastric cancer in Alaska Native people" which sounds considerably prospective and a promising one, but certainly indicates a totally different kind of study.*

Response to Reviewer comment about study design and IRB protocol: Under the IRB we proposed a two part study. The first part was to conduct an in depth analysis of the clinicopathological data which we are presenting in this first paper. The second part that is currently ongoing and will be incorporated into a second paper, is to perform molecular characterization of paraffin embedded tissue samples from patients included in the first part of the study. The authors believed that the amount of data generated in the molecular characterization study would be too large to include in combination with the epidemiological data. Therefore, we have broken the study under one IRB into two parts, first publishing the epidemiological data and then the molecular characterization data.

Reviewer Three Major Comment: *On the other hand, this manuscript has severe limitations, considering the small number of AN patients and the slow aggregation rate do NOT provide adequate sample in order to conduct safe investigation of all the confounders.*

Response to Reviewer comment about slow aggregation rate and adequate sample: The Reviewer brings up an important point that was also addressed in Reviewer one and two comments. We understand the number of AN people included in this study, 132 individuals, is small. In order to conduct this study, we identified all AN gastric cancer patients diagnosed from 2006-2014 at the Alaska Native Medical Center (ANMC). ANMC is the only state-wide referral hospital that oversees specialty care such as,

medical oncology therefore all AN cancer patients receive their oncology care at ANMC or elect to receive their care at a private hospital. To further address this comment, we have added the following sentence to the discussion:

> The AN population is relatively small- consisting of 150,000 people. In order to conduct this study, we reviewed all AN gastric cancer cases diagnosed at the Alaska Native Medical Center between 2006-2014, approximately 132 cases. Records from this timespan had the epidemiological information needed to conduct this study, which is why we focused on these individuals. Even with the small number of cases, we were able to detect significant differences in the results. Although the AN population is small, we feel this population is worthy of study because of the poor clinical outcomes and gastric cancer mortality rates that are unique to this population within Alaska.

Reviewer Three Major Comment: *Additionally, there is a loss of data from history due to retrospective nature of this report, and data missing for about 20% of all AN gastric cancer patients, which introduces a serious element of selection bias.*

Response to Reviewer comment on selection bias and 20% of missing data: We agree with the Reviewer that a limitation of our study is that 20% of AN patients are not accounted for in the ANMC hospital registry. The patients not included in the hospital registry may receive care within the state or outside the state at private hospitals due to convivence in location or additional treatment options available at these hospitals. For example, Southwest Alaska communities are closer to the Pacific Northwest hospitals therefore patients may choose to travel to Seattle, WA instead of Anchorage, AK for their care. Also, patients may choose to participate in a clinical trials offered at hospitals outside of Alaska, because there are no clinical trials are currently offered at ANMC. ANMC is the only state-wide referral hospital that serves all of the AN people through the Indian Health Service (IHS). One of the main reasons for utilizing the ANMC

hospital registry from 2006-2014 was the availability of patient epidemiological and clinical outcome data and access to electronic patient medical records that were not available through the SEER registry. We have included SEER registry data on AN people in Table 1 to show that similar trends observed in our AN ANMC hospital registry data are also represented in the AN SEER registry data. Furthermore, there were no significant differences in clinical or pathological characteristics between the AN SEER and AN ANMC hospital-based registry data. To address the Reviewer's comment on selection bias and 20% of missing data, we added a sentence to the results and discussion:

Methods:

> The SEER database captures all cancer cases among the AN population, approximately 150,000 people, through the Alaska Native Tumor Registry.

Results:

> Similar trends were observed between the AN Hospital and AN SEER data.

Discussion:

> It is possible that by not including all AN people we are introducing an element of selection bias into our results, however no significant differences in clinical or pathological characteristics were seen between the AN SEER and AN ANMC Hospital-based registries. By utilizing the ANMC hospital-based registry we were able to further evaluate clinicopathological and treatment outcomes that are not collected by the SEER registry.

REVIEWER FOUR COMMENTS TO AUTHORS

Reviewer Four Major Comment: *The only difference of gastric cancer between the East Asian and Alaskan Native people was age distribution. The prevalence rate of gastric cancer is higher in the elder people, however, this manuscript pointed out the high prevalence in the under 54 years in Alaskan Native people. The author can explain the mechanisms for this phenomenon.*

Response to Reviewer comment regarding differences between Eastern Asian and Alaska Native patients that may drive younger age at diagnosis: Reviewer one also asked us to explain why the Alaska Native (AN) people are diagnosed at an earlier age compared to the non-Hispanic White population. There could be multiple reasons as to why the AN people are younger age at time of diagnosis of gastric cancer. Firstly, based on extensive understanding of the promotional role of *Helicobacter pylori* in gastric cancer, it is widely known that exposure to *H. pylori* is primary responsible for the increased incidence, non-cardia location, and possibly earlier age of diagnosis. In a study performed by the Centers for Disease Control, they determined AN children were infected with *H. pylori* early in life, often by the age of 4. This may be similar to the Eastern Asian population whom also has a higher incidence of *H. pylori* infection.

One difference between the Eastern Asian populations and the AN population is the higher prevalence of diffuse type in particular signet ring cell carcinomas among AN patients, 22% Eastern Asian to 39% AN of total gastric cancer cases. Diffuse type and signet ring cell carcinomas have been associated with a younger age distribution and female predominance, which is similar to what we've observed in our study. However, in our study there were no significant differences in sex, stage, age at diagnosis, or overall survival between signet ring cell carcinoma and non-signet ring cell carcinoma AN patients. One factor that may drive the high incidence of diffuse and signet ring carcinomas is chronic inflammation through autoimmune gastritis or

chronic *H. pylori* infection. Little research has been conducted to determine whether AN people have a higher incidence of autoimmune gastritis compared to other populations. However, the AN people have a very high prevalence rate of *H. pylori,* 75% of AN people are seroprevalence positive. We are currently investigating the role of chronic inflammation in the promotion of AN gastric cancers to identity alternative biomarkers for earlier detection and treatment of this devastating disease.

We have also included in the discussion other factors that could play a role in younger age at diagnosis such as early exposure to tobacco, genetic predisposition, and early age of diagnosis of other cancer types among AN people. We have added a paragraph to the discussion on possible reasons of early age of onset.

> The younger age at diagnosis among AN patients with gastric cancer could be driven by multiple etiologies. One factor is earlier exposure to particular gastric cancer risk factors such as *H. pylori* infection and tobacco use. Previous research revealed 40% of AN children have been infected with H. pylori by age 4, 70% by age 10, and 78% by age 14[24]. This study and our results suggest the likelihood of long term exposure to systemic inflammation due to the early age of acquisition of *H. pylori* may play an important role in the high incidence of non-cardia cancer, younger age at diagnosis, and the overall gastric cancer health disparity among the AN people. Further, the high prevalence of tobacco use among the AN people may also contribute to the younger age of diagnosis of gastric cancer patients. Another variable associated with gastric cancer in younger individuals is genetic predisposition such as CDH1 germline mutations that result in hereditary diffuse gastric cancers. Approximately 30% of AN patients had a family history of gastrointestinal cancers and there was no difference in age of diagnosis. Further, other types of cancer among the AN people such as lung, kidney, and colorectal cancer are also associated with younger age of diagnosis suggesting earlier age of diagnosis of cancer is a general characteristic in AN patients compared to NHW. Often cancers diagnosed at a younger age are more

aggressive and are found at a later stage, which may also contribute to cancer health disparities among the AN people.

The following Article Highlights were added to the manuscript:

ARTICLE HIGHLIGHTS

Research background

Gastric cancer is a leading cancer health disparity among the AN people, with a 3-fold higher incidence and mortality rate compared to U.S. NHW people. There are currently a paucity of studies investigating the clinicopathologic features of this disease in AN people, and their relationship to clinical outcomes.

Research motivation

This study was conducted to gain a deeper understanding of AN gastric cancer patient characteristics, pathologic variables, clinical patterns of care, and patient outcomes to gain insights into to this cancer health disparity.

Research objectives

In order to further investigate how to reduce gastric cancer incidence and mortality rates among the AN population, we sought to evaluate recent trends in gastric cancer incidence, response to treatment, and overall survival outcomes in this high incidence population. A greater understanding of gastric cancer incidence and response to treatment among the AN people may facilitate the design of screening programs or the identification of early detection measures, and elucidate new areas for future investigation to potentially reduce incidence and improve patient outcomes.

Research methods

We performed a retrospective analysis of 132 AN gastric cancer patients treated at the Alaska Native Medical Center (ANMC) from 2006-2014, utilizing the ANMC Tumor

Registry and manual patient chart reviews. We compared our findings to data on US NHW and AN gastric adenocarcinoma patients obtained from the US National Institute's SEER Program of the National Cancer Institute 18 dataset for the period 2006-2014. Data were analyzed using software SPSS 23.0.

Research results

AN patients differ from NHW patients in that they have a higher prevalence of non-cardia tumors, unique histological features with a higher incidence of the diffuse subtype, and a higher incidence of signet ring cell carcinomas. AN females were more likely to be diagnosed with stage IV cancers compared to AN males. We observed a decreased overall survival among AN patients with advanced stage disease, O+ blood type, <15 lymph nodes examined at resection, and no treatment. AN gastric cancer patients have a higher incidence rate, a poorer overall survival, and are diagnosed at a significantly younger age compared to NHW patients. This study is the first report detailing the clinicopathologic features of gastric cancer in AN people, as well as information on patterns of care, and clinical outcome data.

Research conclusions

Gastric cancer in AN people is distinct from the NHW population. AN patients were observed to have increased incidence, poorer prognosis, earlier age of diagnosis, and variation in location, and histological subtype of gastric cancer. These clinicopathological characteristics could be driven by multiple variables including, socioeconomic factors and biological differences, such as lifestyle differences, genetic alterations, and environmental exposures. Our findings confirm the importance of early detection, treatment, and surgical resection for AN patients with resectable gastric adenocarcinoma in order to optimize patient outcomes. This study highlights the need for further investigation into understanding the basis for the increased incidence and poorer prognosis of this devastating cancer in AN people.

Research perspectives

Our work highlights the unique clinical and pathologic features of gastric cancer in the AN population. The high incidence of this cancer warrants prompt referral for endoscopic evaluation of AN patients presenting with gastrointestinal symptoms. Of particular concern is the finding that younger women present more frequently with stage IV disease, emphasizing the need to consider a diagnosis of gastric cancer earlier in this population. Clinical outcomes are poor in this population, despite the fact that patients are treated according to standard guidelines. An important area for future study will be investigations into the molecular features of gastric cancer in AN people, with the goal of identifying new prognostic and predictive markers that may improve treatment regimens, and possibly identify new targets for precision medicine.