GPNMB expression impacts prognosis and immune infiltration in cancers

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Primary research

Keywords: GPNMB, immune gene markers, prognosis, tumor-infiltrating

Posted Date: July 20th, 2020

DOI: <https://doi.org/10.21203/rs.3.rs-41771/v1>

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Abstract

Background

Glycoprotein non-metastatic gene B (GPNMB) can regulate tumor progression by interacting with T cell function. However, the association between GPNMB and tumor-infiltrating immune cells and prognosis of various cancers is poorly understood.

Methods

We use the Oncomine and TIMER database to investigate GPNMB expression in multiple tumors. The PrognoScan database, Kaplan-Meier plotter are used to analyze tumor prognosis of GPNMB. R packages are used to performed multivariable cox regression analysis. We use TIMER and GEPIA database to explore the association between GPNMB expression and tumor immune infiltration levels, and immune cell markers. GPNMB related transcription factors and transcription-target networks are investigated via TTRUST database and GeneMANIA .

Results

A high level of GPNMB expression was significantly associated with poor prognosis in stomach adenocarcinoma (STAD). While, a high level of GPNMB expression was significantly associated with favorable prognosis in lung adenocarcinoma (LUAD). Besides, GPNMB expression levels can impact the prognosis in STAD and LUAD patients with lymph node metastasis. Moreover, GPNMB expression level has significant relationships with B cells, CD8 + T cells, CD4 + T cells, macrophages, neutrophils, and DCs infiltrating levels in STAD and LUAD. Besides, various immune gene markers of STAD and LUAD are significantly related to GPNMB expression. In addition, the GPNMB related transcription factors are MITF and TP53. The transcript-target networks are mainly responsible for signal transduction in response to DNA damage, DNA damage response, signal transduction by p53 class mediator, mitotic G1 DNA damage checkpoint, G1 DNA damage checkpoint.

Conclusions

These results indicate that GPNMB is significantly associated with prognosis and immune infiltrating levels in various cancers patients, especially in STAD, LUAD patients. Multiple immune gene markers of STAD and LUAD are significantly related to GPNMB expression, especially monocyte, macrophage polarization, and functional T cells gene markers. Our study signifies that GPNMB plays an essential role in prognosis prediction and immune infiltration of STAD and LUAD.

Background

Cancer-related deaths are one of the critical events in the world. The conventional treatments for cancers are chemotherapy, target therapy, radiotherapy, and surgery. However, the 5-year survival status of cancer patients remains to be unexciting. There are various immune cell subpopulations in the solid tumor microenvironment, and they act as pro-tumorigenic roles or anti-tumorigenic roles by activating or inhibiting some kinds of immune cells[1-3]. Recently, immunotherapy plays a more and more critical role in cancers treatment such as adoptive cellular therapy, inhibitory antibodies of PD-1, and PD-L1, CTLA-4 [4-5]. The high density of effector T cells in solid tumors microenvironment is related to favorable prognosis[6]. Besides, tumor-infiltrating immune cells, including macrophages, Dendritic cells(NK cells), neutrophil granulocytes, can impact cancer immunotherapy efficacy [7]. The cancer immunotherapy is still in the early-stage[8]. The relationship between various cancers and microenvironment immune cells is poorly understood. Therefore, it is essential to find favorable immune-related biomarkers in various cancers types.

GPNMB is called glycoprotein non-metastatic gene B (NMB) and locates on the small arm of chromosome 7 (7p15), which express in multiple cancers and cells, including osteoblasts, macrophages and dendritic cells [9-10]. A study reported that knock-down of GPNMB would promote inflammatory cytokines IL-1 β and TNF- α secretion by regulating M1, M2 polarization[11-14]. Another study reported that GPNMB could make inflammatory hepatic cells transfer to hepatocellular cancer in the process of chronic inflammation state[15]. Besides, GPNMB of infiltrating immune cells such as macrophages and dendritic cells, can inhibit T-cell activation or activate naive T-cells[16-19]. Also, GPNMB was detected highest in dendritic cells of antigen-presenting cells (APC), and it can bind to syndecan-4 of activated T cells to reduce T cells activation[20].

Furthermore, GPNMB can function as a tumor suppressor or tumor activator in cancers[21]. High expression of GPNMB was related to high risk of death in glioblastoma and lung cancer[22-23]. Besides, GPNMB can promote breast cancer or melanoma progression and metastasis by regulating T cells in the tumor microenvironment [24-26]. Interestingly, it was reported that GPNMB was a tumor suppressor gene in colon adenocarcinoma, and overexpression of GPNMB could inhibit colon cancer proliferation, invasion, and migration[27]. One study reported that anti-GPNMB mAb in the tumor microenvironment could inhibit colon cancer progression through increasing T cells [28]. These results indicated that GPNMB was associated with tumor infiltrating immune cells such as macrophages, T cells, and dendritic cells. However, the mechanisms between GPNMB and tumor immunology is not fully understood.

In our study, we used Oncomine, PrognoScan, and Kaplan-Meier plotter to explore whether GPNMB expression impacts tumor prognosis. Besides, we used TIMER to explore the association between GPNMB and tumor-infiltrating immune cells in various cancer types. Our study puts emphasis on the correlation between GPNMB expression and STAD, LUAD.

Methods

ONCOMINE database

ONCOMINE database (www.oncomine.org) is an online cancer microarray database, including DNA or RNA sequences analysis [29]. In our study, we collect GPNMB transcription expressions of different cancer tissues and adjacent normal tissues from the ONCOMINE database. We use p-value: 0.001, fold change: 1.5, gene rank: 10%, data type: mRNA as cut-off of p-value and fold change.

Timer

TIMER (<https://cistrome.shinyapps.io/timer/>) from TCGA, is an open website to investigate the correlation between genes and immune cell infiltration[30]. In our study, we use the “Gene module” to estimate the relationship between GPNMB expression and immune cell infiltration. Spearman's relation and statistical significance are used to evaluate the correlation between GPNMB expression and tumor-infiltrating immune gene markers. Moreover, the correlation standard is evaluated using the value: very weak (0.00–0.19), weak (0.20–0.39), moderate (0.40–0.59), strong (0.60–0.79), very strong (0.80–1.0). P-value < 0.05 is considered to be significantly different.

Prognoscan

Prognoscan database (<http://www.abren.net/Prognoscan/>) is a cancer microarray datasets that can be used to investigate the relationship of GPNMB expression and survival prognosis in multiple cancers types with a Cox P-value< 0.05 [31].

Kaplan-Meier Plotter

We used the Kaplan Meier plotter (<http://kmplot.com/analysis/>) to analyze the prognostic value of GPNMB expression in cancers such as breast, ovarian, lung, liver, and gastric cancer samples[32]. In Kaplan-Meier plotter, patients are divided into high and low expression group according to median values of mRNA expression and KM survival curves is used to validate the survival status. P-value < 0.05 is considered to be significantly different.

GEPIA Dataset

GEPIA (<http://gepia.cancer-pku.cn/index.html>) provides mRNA expression data of tumors and normal samples according to Cancer Genome Atlas (TCGA) and Genotype tissue Expression dataset projects [33]. In this study, we use GEPIA to confirm the relationship between GPNMB expression and immune gene markers in TIMER.

TRRUST

TRRUST (<https://www.grnpedia.org/trrust/>) is an open website to investigate transcript regulatory networks, and it contains 8444 transcription factor regulatory relationships[34].

GeneMANIA

GeneMANIA (<http://www.genemania.org>) is a web that could construct a protein interaction (PPI) network, gene interaction, co-expression, gene enrichment, and gene co-localization[35]. In this study, we use GeneMANIA to construct GPNMB related transcription factor-target network.

Statistical analysis

Multivariable Cox regression analysis was conducted with R package (version 3.6.3) using a significant level of 0.05.

Results

GPNMB mRNA expression in Different Cancer Types

We used the oncomine database (www.oncomine.org) to explore the expression of GPNMB expression in tumors and normal tissues of various cancer types. This study revealed that the GPNMB expression was higher in brain and CNS cancer, cervical cancer, esophageal cancer, gastric cancer, head and neck cancer, kidney cancer, liver cancer, lung cancer, leukemia, lymphoma, melanoma, pancreatic cancer, prostate cancer, and sarcoma compared to normal tissues (**Figure 1A**). Moreover, lower expression was detected in bladder, breast, colorectal, esophageal cancer, leukemia, lung, ovarian, and sarcoma cancers in some data sets. We summarized the detailed results of GPNMB expression in various cancer types in **table I**. Then, TIMER was used to further explore GPNMB expression in various cancer types and normal tissues in Figure 1B. GPNMB expression was significantly lower in BLCA (bladder urothelial bladder carcinoma), BRCA (invasive breast carcinoma), COAD (colon adenocarcinoma), READ (rectum adenocarcinoma), LUAD (lung adenocarcinoma), and UCEC (uterine corpus endometrial carcinoma) compared to adjacent healthy tissues. Besides, GPNMB expression was significantly higher in CHOL (cholangiocarcinoma), ESCA (Esophageal carcinoma), HNSC (head and neck cancer), KICH (kidney chromophobe), KIRC(kidney renal clear cell carcinoma), KIRP(Kidney renal papillary cell carcinoma), LIHC (liver hepatocellular carcinoma), LUSC (Lung squamous cell carcinoma), STAD(stomach adenocarcinoma) compared to adjacent normal tissues.

Potential Prognosis of GPNMB in Cancers

We used Prognoscan to explore the relationship between GPNMB expression and survival status of various cancer types in Figure 2. The associations between GPNMB expression and different cancer types prognosis from Prognoscan were displayed in Figure 3. Patients with high GPNMB expression had poor prognosis in colorectal, breast, skin cancer, prostate cancers (**Figures 2/3**). Patients with high GPNMB expression had favorable prognosis in lung cancer

(Figures 2/3). In addition, we used Kaplan-Meier plotter database to further explore whether GPNMB impacts the prognosis in various cancer types. The results showed that higher level of GPNMB expression was associated with poor prognosis in stomach adenocarcinoma (OS HR= 1.59, 95% CI =1.11-2.27, P = 0.011; RFS HR = 2.61, 95% CI = 1.01-6.67, P = 0.039), LIHC cancer (OS HR= 1.83, 95% CI = 1.27-2.66, P = 0.0011), breast cancer (OS HR= 1.8, 95% CI = 1.27-2.56, P = 0.00082), and ovarian cancer (OS HR= 1.15, 95% CI = 1.01-1.31, P = 0.034; PFS HR = 1.35, 95% CI = 1.19-1.53, P = 4e-6) (Figures 2I-P). The results showed that higher level of GPNMB expression was associated with favorable prognosis in lung adenocarcinoma(OS HR=0.7, 95%CI=0.51-0.96, P=0.028, Figures 2N)

GPNMB expression influences the prognosis of gastric cancer and lung adenocarcinoma patients with clinical characteristics

We used Kaplan-Meier plotter databases to explore whether GPNMB expression was related to clinical characteristics in gastric cancer patients. Overexpression of GPNMB was related to favorable OS in male patients. Overexpression of GPNMB was related to favorable PPS in male and female patients (P < 0.05). Specifically, overexpression of GPNMB was associated with favorable OS in stage 1 but was related to poor PPS of stage 2 (OS HR = 0.27, P = 0.0054; PPS HR =0.52, P = 0.048) in gastric cancer patients. Overexpression of GPNMB was associated with favorable OS in T3 stage and favorable PPS in T3-4 stage of gastric cancer patients (OS HR = 0.69, P = 0.035; PPS HR = 0.6, P = 0.014; PPS HR = 0.24, P = 0.016). Interestingly, high expression of GPNMB was associated with poor OS in M0 stage (OS HR =1.42, P = 0.015). High expression of GPNMB was associated with poor OS in N1, N1+2+3 stage and poor PPS in N1, N2, N1+2+3 stage (OS HR =1.42, P = 0.015; OS HR =1.5, P = 0.0038; PPS HR =1.81, P = 0.013; PPS HR =0.5, P = 0.0086; PPS HR =1.58, P = 0.003). Overexpression of GPNMB was associated with poor OS in intestinal and diffuse Lauren classification but favorable OS in mixed Lauren classification (OS HR =1.58, P = 0.0042; OS HR =1.5, P = 0.019; OS HR =0.21, P = 0.0022). Overexpression of GPNMB was associated with poor PPS in diffuse Lauren classification (PPS HR = 1.9, P = 0.0019). High expression of GPNMB was correlated with poor OS in well and moderate differentiated type but favorable OS in poorly differentiated type (OS HR =0.59, P = 0.011; OS HR =2.41, P = 0.0083; OS HR =6.43, P = 0.0044). Besides, high expression of GPNMB was correlated with favorable PPS in poorly differentiated type (PPS, HR =0.49, P = 0.039) (Table II). These results indicate that GPNMB expression level is related to prognosis in gen-der, stage1-2, T3-4 stage, N1-3 stage, M0 stage, Lauren classification and differentiation of gastric cancer patients.

In addition, we enrolled 410 LUAD patients from TCGA to explore the prognostic value of GPNMB and other clinical factors. The multivariable Cox regression analysis indicates that the OS of LUAD patients is significantly associated with GPNMB expression, T stage, N stage and stage (all P<0.05, figure 4).

Specifically, N stage signifies regional lymph node metastasis, and the present study reveal that GPNMB expression level can impact the prognosis in STAD, and LUAD patients with lymph node metastasis.

GPNMB expression is associated with immune infiltration level in cancers

We used TIMER to explore the association between GPNMB expression and immune infiltration levels in 39 cancer types. Our study reveals that GPNMB expression is significantly related to tumor purity in 27 cancer types and significantly related to B cell infiltration levels in 25 cancer types. Besides, GPNMB expression is significantly related to CD8+ T cells infiltrating levels in 21 cancer types, CD4+ T cells infiltrating levels in 23 cancer types, macrophages in 29 cancer types, neutrophils in 28 cancer types, and dendritic cells in 32 cancer types.

Then, we further investigated the relationship between GPNMB expression and immune infiltration in distinct cancers. Interestingly, our study reveals that GPNMB expression is significantly related to high immune infiltration level in Breast, COAD, LUAD, LIHC, STAD. GPNMB expression level had significant positive relationships with B cell (r =0.250 , P = 1.92E-15), CD8+ T cells (r =0.471 , P = 4.43E-55), CD4+ T cells (r = 0.379, P = 3.28E-34), macrophages (r = 0.581, P = 7.05E-90), neutrophils (r = 0.547, P = 2.655E-75) and DCs (r =0.569, P = 1.34E-82) infiltrating levels in BRCA (Figure 5A). GPNMB expression level had significantly positive relationships with B cell (r =0.200 , P = 4.56E-5), CD8+ T cells (r =0.382 , P = 1.51E-15), CD4+ T cells (r = 0.443, P = 1.01E-20), macrophages (r = 0.746, P = 4.70E-73), neutrophils (r = 0.663, P =3.90E-52) and DCs (r =0.727, P = 2.88E-67) infiltrating levels in COAD (Figure 5B). GPNMB expression level had significant positive relationships with B cells (r =0.492 , P = 2.35E-22), CD8+ T cells (r =0.535 , P = 1.10E-26), CD4+ T cells (r = 0.397, P = 2.03E-14), macrophages (r = 0.629, P = 6.50E-39), neutrophils (r = 0.542, P =9.76E-28) and DCs (r =0.678, P = 3.57E-47) infiltrating levels in LIHC (Figure 5C). GPNMB expression level had significant positive relationships with B cells (r =0.287, P = 1.30E-10), CD8+ T cells (r =0.339 , P = 1.41E-14), CD4+ T cells (r = 0.309, P = 3.81E-12), macrophages (r = 0.593, P = 2.41E-47), neutrophils (r = 0.585, P =1.26E-45) and DCs (r =0.697, P = 3.13E-72) infiltrating levels in LUAD (Figure 5D). In addition, GPNMB expression had no significant correlations with tumor purity and CD4+ T cells, macrophages infiltrating levels in LUSC (Figure 5E). GPNMB expression level had significantly negative relationship with B cell (r =-0.105 , P = 0.0225) , CD8+ T cells (r =-0.122 , P = 7.77E-3), neutrophils (r = -0.113, P =1.36E-2) and DCs (r =-0.11, P = 0.0166) infiltrating levels in LUSC but the relationships were weak (Figure 5E). GPNMB expression level had significant positive relationship with CD8+ T cells (r =0.487 , P = 2.14E-23), CD4+ T cells (r = 0.272, P = 1.3E-7), macrophages (r = 0.657, P = 4E-47), neutrophils (r = 0.621, P =5.62E-41) and DCs (r =0.761, P = 2.83E-71) infiltrating levels and had significant negative relationship with B Cell infiltrating levels (r =-0.261 , P = 3.52E-7) in STAD (Figure 5F). The results above displayed that GPNMB is tightly related to immune infiltration in Breast, COAD, LUAD, LIHC, STAD.

The relationship between GPNMB expression and immune markers

T cells are comprised of T cells CD8+ lymphocytes, helper T cells, memory T cells, and T regulatory cells (FOXP3+). And functional T cells are comprised of T helper cells, Treg, and T cell exhaustion. Our study used TIMER and GEPIA databases to investigate the relationship between GPNMB and immune cells immune markers included CD8+ T cells, T cells (general), B cells, monocytes, TAMs, M1 and M2 macrophages, neutrophils, NK cells, DCs, functional T cells and exhausted T cells in STAD and LUAD with the control of LUSC (Table III and Figure 6). Gene markers of monocyte such as CD86 and CD115 have strong relationships with GPNMB expression, and gene markers of TAM such as CCL2, CD68, IL10 have a moderate or weak correlation with GPNMB, and M1 macrophages gene markers such as IRF5 has a weak association with GPNMB expression, and gene markers of M2 macrophages such as CD163, VSIG4, MS4A4A have a moderate and strong association with GPNMB expression in STAD, and LUAD (Tables III, IV). Our study indicated that GPNMB expression level was significantly related to most of the immune cell's immune markers in STAD, and LUAD, especially monocytes, TAMs, M1, and M2 macrophages markers. Nevertheless, the GPNMB expression level was significantly associated with only 20 immune markers in LUSC (Table III). Specially, we revealed CD86,

CD115 of Monocyte, CCL-2, CD68, IL10 of TAMs, IRF5 of M1 Macrophage, CD163, VSIG4 and MS4A4A of M2 phenotype are significantly correlate with GPNMB expression in STAD and LUAD ($P < 0.05$; Figure 6A–L). We further used the GEPIA database to verify the relationship between GPNMB expression and the immune markers of monocytes, TAMs, M1, and M2 macrophages markers in STAD, LUAD and LUSC, and the results were similar to those of TIMER (Table IV). Our results indicate that GPNMB is associated with macrophage polarization in STAD and LUAD.

Besides, GPNMB expression has a positive relationship with the DC infiltration level in STAD, and LUAD. DC immune markers such as HLA-DPB1, HLA-DQB1, HLA-DRA, HLA-DPA1, BDCA-1, BDCA-4, and CD11c are significantly related to GPNMB expression. In addition, our study signified that GPNMB had a positive relationship with T helper cells (Th1, Th2, Tfh, and Th17), Treg and T cell exhaustion immune markers such as T-bet, STAT4, STAT1, IFN- γ , TNF- α , GATA3, STAT5A, IL13, BCL6, IL21, STAT3, FOXP3, CCR8, STAT5B, TGF β , PD-1, CTLA4, LAG3 and TIM-3 in STAD, and LUAD ($P < 0.0001$, Table III). In summary, our study signifies that GPNMB plays an essential role in the immune microenvironment of STAD, and LUAD.

Enrichment analysis of GPNMB related transcription factors functional networks

TTRUST database revealed that GPNMB related transcription factors were MITF and TP53 (Table V). Also, we used GeneMANIA to construct the transcription-target network (figure 7). The functional analysis of the transcription-target network is mainly responsible for signal transduction in response to DNA damage, DNA damage response, signal transduction by p53 class mediator, mitotic G1 DNA damage checkpoint, G1 DNA damage checkpoint.

Discussion

GPNMB is a type I transmembrane glycoprotein and correlated with cancer growth and metastasis. GPNMB elevated in infiltrating immune cells such as macrophages, dendritic cells, CD14+ monocytes [16-19]. Besides, GPNMB can elevate T cells in the tumor microenvironment to promote cancer progression and metastasis [24-26]. In the present study, we reveal the association between GPNMB expression and various cancers prognosis. A higher level of GPNMB expression was associated with poor prognosis in gastric cancer. A higher level of GPNMB expression was associated with favorable prognosis in lung cancer. Also, our study signifies that GPNMB expression is associated with tumor infiltration level and immune cell markers of gastric cancer and lung adenocarcinoma.

In the present study, we used Oncomine and TIMER to explore the GPNMB expression in various cancer types and normal tissues. According to the oncomine database, GPNMB expression elevated in brain and CNS cancer, breast cancer, cervical cancer, esophageal cancer, gastric cancer, head and neck cancer, kidney cancer, leukemia, liver cancer, lung cancer, lymphoma, melanoma, pancreatic cancer, prostate cancer, and stroma; In contrast, GPNMB expression reduced in bladder, breast, colorectal, esophageal cancer, leukemia, lung, ovarian, and sarcoma cancers in some data sets (Figure 1A). Based on TIMER which includes TCGA data, GPNMB expression significantly reduced in BLCA, BRCA, COAD, READ, LUAD, and UCEC compared to adjacent healthy tissues; while GPNMB expression elevated in CHOL, HNSC, KICH, KIRC, KIRP, LUSC, LIHC, and STAD compared to adjacent normal tissues (Figure 1B). Then, we used PrognoScan and Kaplan-Meier Plotter to analyze the correlation between GPNMB expression and cancer prognosis. PrognoScan showed that high GPNMB expression was significantly associated with poor prognosis in blood, brain, colorectal, breast, skin cancer, prostate cancers (Figures 2/3). Besides, the Kaplan Meier plotter database indicated that a higher level of GPNMB expression correlated with poor prognosis in stomach adenocarcinoma, LIHC cancer, breast cancer, and ovarian cancer, and correlated with favorable prognosis in lung adenocarcinoma. In summary, our study reveals that GPNMB could be a prognostic biomarker in blood, brain, colorectal, breast, LUAD, melanoma, prostate, ovarian, LIHC, and gastric cancer.

Besides, low expression of GPNMB is associated with prognosis in gender, stage1-2, T3-4 stage, N1-3 stage, M0 stage, Lauren classification, and differentiation of gastric cancer (Table II). The multivariable Cox regression analysis indicates that the OS of LUAD patients from TCGA is significantly associated with GPNMB expression, T stage, N stage and stage (all $P < 0.05$, figure 4). We can infer that GPNMB expression level can impact the prognosis in STAD and LUAD patients with lymph node metastasis.

Previous studies reported that tumor-infiltrating lymphocytes' density and location were significantly related to sentinel lymph node status and impacted survival status in colorectal cancer and melanoma [36-37]. In the present study, we investigated the correlations between GPNMB expression and tumor-infiltrating immune cells. The results indicate that GPNMB expression was correlated moderately or strongly with macrophages and DC and positively related to CD8+ T, CD4+ T cells, and neutrophils in BRCA, LIHC, LUAD, COAD, and STAD (Figures 3A-D, F). Furthermore, we investigated the association between GPNMB expression and tumor-infiltrating immune cells markers in STAD and LUAD with control of LUSC. Gene markers of monocyte such as CD86 and CD115 had strong relationships with GPNMB expression, and gene markers of TAM such as CCL2, CD68, IL10 had a moderate or weak correlation with GPNMB, and M1 macrophages gene markers such as IRF5 had a weak association with GPNMB expression, and gene markers of M2 macrophages such as CD163, VSIG4, MS4A4A had a moderate and strong association with GPNMB expression (Tables III, IV). All the results suggest that GPNMB expression plays a critical part in regulating tumor-infiltrating monocyte and macrophages in STAD, and LUAD. Moreover, Our study also signifies that GPNMB has a positive relationship with T helper cells (Th1, Th2, Tfh, and Th17), Treg and T cell exhaustion immune markers in STAD, and LUAD ($P < 0.0001$, Table III). In summary, Our study signifies that GPNMB plays a vital role in the immune microenvironment of STAD and LUAD. In addition, we used TTRUST to explore GPNMB related transcription factors, which are MITF and TP53. GeneMANIA was used to construct a transcription-target network, which is mainly responsible for signal transduction in response to DNA damage, DNA damage response, signal transduction by p53 class mediator, mitotic G1 DNA damage checkpoint, G1 DNA damage checkpoint.

In melanoma, breast cancer, gliomas, prostate cancer, lung cancer, and bladder cancer, It was reported that high expression of GPNMB could promote cancer cell progression and invasion, and downregulate cancer cell apoptosis [24-25, 38-43]. In colon cancer, tumor-infiltrating immune cells such as T cells could reduce tumor progression and the density, distribution of immune cells could have advantages in predicting survival status than TNM or Duke's classification [44-45]. In gastric cancer, DC cells recognize and present the tumor neoantigens to the T cells MHC molecule-s; then, effector T cells are activated, subsequently inhibiting gastric cancer cells [46-47]. GPNMB is detected highest in dendritic cells of antigen-presenting cells (APC), and it can bind to syndecan-4 of activated

T cells to reduce T cells activation[20]. The previous study reported that Th2 cells, T helper cell infiltration could produce antitumor immunity and impacted the survival status of cancer patients [48]. Therefore, the correlation between GPNMB and tumor immune infiltration could impact the prognosis in STAD and LUAD.

Conclusions

In summary, our study reveals that GPNMB expression could impact the prognosis of various cancer types such as lung adenocarcinoma, gastric cancer patients, and is positively related to immune cell infiltrating level in various cancer types, especially in STAD, and LUAD. Moreover, GPNMB expression plays a vital part in regulating monocyte, macrophage polarization, and functional T cells in STAD, and LUAD. Therefore, GPNMB could be a potential biomarker for the tumor immune infiltration and prognosis prediction in STAD, and LUAD.

Declarations

Acknowledgment

We sincerely thank the researchers for providing their databases information online, and it is our pleasure to acknowledge their contributions.

Funding

Not applicable

Conflicts of Interest

The authors have no conflicts of interest to declare.

Ethical Statement:

The current study received approval from the Xiangya Hospital of Central South University according to the Declaration of Helsinki. The retrieval of every data was collected from the open web, confirming that all of the written informed consents were attained. The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Author Contributions

Yang Wang conceived, collected, analyzed the data. Qiong Chen conceptualized, developed an outline for the manuscript, revised the manuscript and approved the final version to be published. Yang Wang, Qiong Chen wrote the manuscript and approved the final manuscript.

Consent for publication

This manuscript is approved by all authors for publication

Availability of data and materials

The datasets generated during our study are not publicly available but available on reasonable request.

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Tables

Table I. GPNMB expression in cancers versus normal tissues in ONCOMINE database

Cancer	Cancer type	P-value	Fold change	sample	reference
Bladder cancer	Superficial Bladder Cancer	6.24E-07	-2.146	126	20421545
Brain and CNS cancer	Astrocytoma	1.74E-08	3.141	45	11559565
	Glioblastoma	2.21E-07	4.695	22	16697959
	Glioblastoma	1.14E-06	3.173	27	16204036
	Anaplastic Oligoastrocytoma	0.000773	3.353	4	16357140
Breast cancer	invasive breast carcinoma	1.49E-17	4.038	53	18438415
	Ductal Breast Carcinoma	1.68E-06	2.342	40	16473279
	invasive lobular breast carcinoma	1.02E-20	-1.859	148	22522925
	invasive ductal breast carcinoma	1.42E-31	-1.849	1556	22522925
cervical cancer	Cervical Squamous Cell Carcinoma	7.98E-07	1.585	40	18191186
Colorectal cancer	Colon Adenoma	5.94E-10	-9.538	5	20957034
	Colon Adenoma	3.93E-12	-3.43	25	18171984
	Rectal Adenoma	5.25E-05	-4.025	7	18171984
	Colorectal Adenocarcinoma	1.47E-09	-2.904	45	20957034
Esophageal cancer	Esophageal Squamous Cell Carcinoma	2.70E-14	2.51	53	PMC3086948
	Barrett's Esophagus	0.000674	-1.87	8	15833844
	Barrett's Esophagus	6.02E-05	-1.906	19	16449976
	Barrett's Esophagus	4.72E-11	-7.961	15	21152079
Gastric cancer	Gastric Mixed Adenocarcinoma	4.93E-07	4.155	8	12925757
	Gastric intestinal type adenocarcinoma	9.85E-18	2.596	66	12925757
	Diffuse Gastric Adenocarcinoma	3.55E-07	3.476	13	12925757
	Gastric Cancer	1.31E-05	4.06	12	21132402
	Diffuse Gastric Adenocarcinoma	0.000196	3.823	6	19081245
Head and neck cancer	oral cavity carcinoma	3.38E-05	2.699	4	17510386
	Tonsillar Carcinoma	0.000847	4.394	6	17510386
	Tongue Carcinoma	3.64E-06	3.039	15	17510386
	Tongue Squamous Cell Carcinoma	3.67E-05	2.473	26	18254958
	Tongue Squamous Cell Carcinoma	1.25E-06	2.692	31	19138406
	Tongue Squamous Cell Carcinoma	8.26E-07	2.994	31	15833835
Kidney cancer	Papillary Renal Cell Carcinoma	0.000774	6.77	4	12598325
	Chromophobe Renal Cell Carcinoma	0.000318	11.809	4	19445733
	Papillary Renal Cell Carcinoma	0.000572	6.324	19	19445733
	Hereditary Clear Cell Renal Cell Carcinoma	1.25E-07	3.574	32	19470766
Leukemia	Hairy cell Leukemia	6.23E-08	15.252	16	15778709
	Pro-B Acute Lymphoblastic Leukemia	7.85E-18	-1.798	70	20406941
Liver cancer	Hepatocellular Carcinoma	1.13E-18	10.073	38	19098997
Lung cancer	Squamous Cell Lung Carcinoma	5.24E-10	3.282	27	20421987
	Lung Carcinoid Tumor	1.17E-12	-47.941	20	11707567
	Lung Adenocarcinoma	3.39E-10	-2.036	58	22613842
Melanoma	Benign Melanocytic Skin Nevus	7.07E-05	2.14	18	16243793
	Cutaneous Melanoma	6.11E-05	2.574	45	16243793
Ovarian cancer	Ovarian Serous Adenocarcinoma	1.77E-13	-4.814	43	19486012
Pancreatic cancer	Pancreatic Adenocarcinoma	8.01E-05	4.177	10	12750293
	Pancreatic Ductal Adenocarcinoma	1.71E-14	6.203	39	19260470
	Pancreatic Adenocarcinoma	0.000758	3.452	12	12651607
Prostate cancer	Prostate Carcinoma	2.53E-06	2.723	52	12086878
Sarcoma	Malignant Fibrous Histiocytoma	1.94E-05	5.701	9	15994966
	Leiomyosarcoma	0.000683	4.228	6	15994966
	Myxoid/Round Cell Liposarcoma	5.69E-09	-5.393	20	20601955
Lymphonoma	Diffuse Large B-Cell Lymphoma	1.49E-42	114.445	44	PMC2973325
	Activated B-Cell-Like Diffuse Large B-Cell Lymphoma	1.28E-22	136.629	17	PMC2973325
	Centroblastic Lymphoma	3.45E-28	169.384	28	15778709
	Primary Effusion Lymphoma	2.92E-05	8.807	9	
	Burkitt's Lymphoma	1.07E-05	45.152	17	15778709
	Hodgkin's Lymphoma	6.54E-07	15.291	12	18794340
	Unspecified Peripheral T-Cell Lymphoma	1.20E-17	63.903	28	17304354
others	Malignant Fibrous Histiocytoma	1.94E-05	5.701	9	15994966
	Testicular Seminoma	6.42E-07	5.356	23	14595015
	Seminoma, NOS	9.35E-07	6.928	12	
	Embryonal Carcinoma, NOS	1.86E-06	5.559	15	16424014
	Teratoma, NOS	2.81E-06	6.846	14	16424014
	Uterine Corpus Leiomyoma	8.48E-05	-1.513	50	19622772

TABLE II. Correlation of GPNMB mRNA expression and clinical prognosis in gastric cancer with different clinical factors by Kaplan-Meier plotter.

characteristics	Overall survival			PPS		
	(n = 881)			(n = 498)		
	N	Hazard ratio	P	N	Hazard ratio	P
SEX						
Female	244	0.72(0.5-1.03)	0.074	149	0.57(0.37-0.88)	0.0099
Male	544	0.73(0.57-0.92)	0.0078	348	0.66(0.51-0.85)	0.0015
STAGE						
1	67	0.27(0.1-0.72)	0.0054	31	0.32(0.06-1.67)	0.15
2	140	1.65(0.77-3.56)	0.2	105	2.52(0.98-6.51)	0.048
3	305	0.79(0.59-1.05)	0.1	142	1.28(0.81-2.02)	0.29
4	148	1.24(0.81-1.9)	0.33	104	1.55(0.94-2.58)	0.086
T stage						
1	†			†		
2	241	1.46(0.95-2.24)	0.084	196	1.56(0.99-2.45)	0.052
3	204	0.69(0.49-1.98)	0.035	150	0.6(0.39-0.91)	0.014
4	38	1.63(0.69-3.82)	0.26	29	0.24(0.07-0.84)	0.016
N stage						
0	74	0.44(0.19-1.03)	0.052	41	0.5(0.16-1.59)	0.23
1	225	1.59(1.05-2.41)	0.027	169	1.81(1.13-2.91)	0.013
2	121	1.99(0.99-2.56)	0.052	105	0.5(0.29-0.85)	0.0086
3	76	1.47(0.8-2.69)	0.22	63	1.65(0.88-3.12)	0.12
1+2+3	422	1.5(1.14-1.97)	0.0038	337	1.58(1.16-2.51)	0.003
M stage						
0	444	1.42(1.07-1.87)	0.015	342	1.33(0.98-1.8)	0.07
1	56	0.76(0.41-1.42)	0.39	36	1.57(0.72-3.43)	0.26
Lauren classification						
intestinal	336	1.58(1.15-2.16)	0.0042	192	1.42(0.94-2.15)	0.093
diffuse	241	1.5(1.07-2.12)	0.019	176	1.9(1.26-2.68)	0.0019
mixed	32	0.21(0.07-0.62)	0.0022	†		
differentiation						
poorly differentiated	165	0.59(0.39-0.89)	0.011	49	0.49(0.25-0.98)	0.039
moderately differentiated	67	2.41(1.23-4.73)	0.0083	24	1.89(0.73-4.87)	0.18
well differentiated	32	6.43(1.49-24.83)	0.0044	†		
†: variables missing. PPS: post-progression survival						

TABLE III. Relationship between GPNMB expression and immune cell markers in TIMER.

STAD, stomach adenocarcinoma; LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma; TAM, tumor-associated macrophage; Th, T helper cell; Tfh, Follicular helper T cell; Treg, regulatory T cell; Cor, R value of Spearman's correlation; None, correlation without adjustment. Purity, correlation adjusted by purity. *P < 0.05; **P < 0.001; ***P < 0.0001.

COAD				STAD				LUAD				LUSC		
None		Purity		None		Purity		None		Purity		None		Purit
Cor	P	Cor	P	Cor	P	Cor	P	Cor	P	Cor	P	Cor	P	Cor
0.574	***	0.489	***	0.484	***	0.469	***	0.34	***	0.232	***	-0.165	**	-0.188
0.328	***	0.271	***	0.316	***	0.306	***	0.293	***	0.203	***	-0.155	**	-0.173
0.51	***	0.394	***	0.45	***	0.434	***	0.365	***	0.251	***	-0.165	**	-0.2
0.603	***	0.508	***	0.442	***	0.429	***	0.419	***	0.315	***	-0.115	*	-0.146
0.603	***	0.518	***	0.541	***	0.537	***	0.443	***	0.348	***	-0.13	*	-0.163
0.374	***	0.253	***	0.25	***	0.235	***	0.258	***	0.134	*	-0.068	0.131	-0.082
0.44	***	0.305	***	0.317	***	0.283	***	0.25	***	0.133	*	-0.066	0.138	-0.08
0.893	***	0.866	***	0.79	***	0.789	***	0.745	***	0.714	***	-0.026	0.561	-0.058
0.82	***	0.782	***	0.777	***	0.77	***	0.682	***	0.644	***	0.033	0.459	0.01
0.74	***	0.692	***	0.48	***	0.448	***	0.482	***	0.426	***	-0.003	0.943	-0.027
0.684	***	0.65	***	0.67	***	0.65	***	0.722	***	0.689	***	0.125	*	0.116
0.694	***	0.672	***	0.655	***	0.645	***	0.624	***	0.567	***	-0.042	0.347	-0.069
-0.054	0.246	-0.125	*	0.074	0.131	0.064	0.211	0.193	***	0.136	*	0.061	0.176	0.055
0.378	***	0.395	***	0.425	***	0.424	***	0.509	***	0.47	***	0.176	**	0.179
0.323	***	0.257	***	0.104	*	0.106	*	-0.044	0.322	-0.064	0.158	0.008	0.852	-0.003
0.884	***	0.858	***	0.787	***	0.78	***	0.697	***	0.66	***	0.019	0.664	0.002
0.848	***	0.822	***	0.757	***	0.746	***	0.688	***	0.661	***	-0.04	0.371	-0.064
0.887	***	0.866	***	0.86	***	0.855	***	0.733	***	0.702	***	-0.02	0.579	-0.049
-0.126	*	-0.084	0.089	0.046	0.348	0.059	0.251	0.227	***	0.227	***	-0.101	*	-0.113
0.846	***	0.828	***	0.697	***	0.686	***	0.69	***	0.665	***	0.042	0.351	0.02
0.496	***	0.385	***	0.402	***	0.386	***	0.417	***	0.32	***	-0.077	0.085	-0.0489
0.29	***	0.231	***	0.298	***	0.309	***	0.079	0.074	0.029	0.517	0.042	0.351	-0.036
0.322	***	0.2561	***	0.259	***	0.249	***	0.18	***	0.11	*	-0.039	0.379	-0.053
0.361	***	0.265	***	0.217	***	0.198	**	0.147	**	0.072	0.1082	-0.088	*	-0.104
0.368	***	0.288	***	0.256	***	0.255	***	0.15	**	0.102	*	-0.032	0.469	-0.055
0.356	***	0.292	***	0.297	***	0.281	***	0.205	***	0.13	*	-0.113	*	-0.127
0.135	*	0.107	*	0.016	0.749	0.015	0.773	0.022	0.62	0.001	0.975	-0.073	0.104	-0.076
0.292	***	0.253	***	0.185	**	0.174	**	0.194	***	0.145	*	-0.050	0.264	-0.052
0.758	***	0.701	***	0.607	***	0.591	***	0.548	***	0.491	***	-0.052	0.256	-0.087
0.519	***	0.43	***	0.416	***	0.39	***	0.427	***	0.364	***	-0.028	0.526	-0.03
0.72	***	0.654	***	0.572	***	0.56	***	0.57	***	0.515	***	-0.096	*	-0.123
0.76	***	0.7	***	0.571	***	0.556	***	0.587	***	0.536	***	-0.081	0.07	-0.105
0.487	***	0.39	***	0.414	***	0.394	***	0.313	***	0.255	***	0.047	0.297	0.035
0.811	***	0.772	***	0.631	***	0.624	***	0.177	***	0.157	**	0.113	*	0.1
0.857	***	0.855	***	0.768	***	0.76	***	0.676	***	0.633	***	0.025	0.575	0.01

0.571	***	0.504	***	0.487	***	0.48	***	0.372	***	0.274	***	-0.098	*	-0.117
0.576	***	0.495	***	0.507	***	0.515	***	0.395	***	0.294	***	-0.058	0.197	-0.075
0.591	***	0.54	***	0.321	***	0.326	***	0.389	***	0.32	***	-0.067	0.133	-0.081
0.429	***	0.375	***	0.325	***	0.328	***	0.269	***	0.178	***	-0.169	0.0001	-0.18
0.427	***	0.384	***	0.427	***	0.149	*	0.345	***	0.262	***	-0.008	0.853	-0.034
0.575	***	0.524	***	0.419	***	0.404	***	0.444	***	0.364	***	-0.019	0.67	-0.033
0.068	0.145	0.073	0.14	0.224	***	0.235	***	0.106	*	0.126	*	0.174	**	0.171
0.381	***	0.365	***	0.543	***	0.538	***	0.556	***	0.497	***	0.023	0.615	0.008
0.347	***	0.292	***	0.141	*	0.141	*	0.082	0.064	0.009	0.834	-0.011	0.805	-0.027
0.591	***	0.516	***	0.394	***	0.383	***	0.123	*	0.111	*	0.205	**	0.211
0.349	***	0.325	***	0.391	***	0.392	***	0.235	***	0.184	***	-0.083	0.064	-0.092
0.403	***	0.345	***	0.451	***	0.45	***	0.199	***	0.206	***	0.156	**	0.15
-0.163	**	-0.199	***	-0.08	0.102	-0.086	0.093	0.148	**	0.085	0.06	-0.146	*	-0.146
0.669	***	0.611	***	0.488	***	0.478	***	0.466	***	0.387	***	-0.013	0.77	-0.038
0.687	***	0.643	***	0.603	***	0.61	***	0.517	***	0.455	***	0.018	0.69	-0.011
0.302	***	0.319	***	0.457	***	0.468	***	0.31	***	0.3	***	0.109	*	0.107
0.760	***	0.697	***	0.501	***	0.487	***	0.376	***	0.306	***	0.218	***	0.224
0.549	***	0.467	***	0.383	***	0.367	***	0.35	***	0.243	***	-0.139	*	-0.164
0.599	***	0.528	***	0.346	***	0.328	***	0.39	***	0.28	***	-0.095	*	-0.129
0.557	***	0.468	***	0.398	***	0.387	***	0.299	***	0.203	***	-0.127	*	-0.153
0.913	***	0.894	***	0.811	***	0.811	***	0.741	***	0.71	***	-0.06	0.18	-0.087
0.197	***	0.163	**	0.354	***	0.329	***	0.262	***	0.154	**	-0.169	**	-0.199

TABLE IV. Relationship between GPNMB expression and immune cell markers in GEPIA.

STAD, stomach adenocarcinoma; LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma. TAM, Tumor-associated macrophages. Tumor, correlation analysis in tumor tissue of TCGA. Normal, correlation analysis in normal tissue of TCGA.*P < 0.05; **P < 0.001; ***P < 0.0001.

Disccription	gene markers	COAD				STAD				LUAD				L	
		Tumor		Normal		Tumor		Normal		Tumor		Normal		Tumor	
		R	P	R	P	R	P	R	P	R	P	R	P	R	P
Monocyte	CD86	0.87	***	0.47	*	0.67	***	-0.35	*	0.57	***	0.71	***	-0.05	0.27
	CD115 (CSF1R)	0.82	***	0.61	***	0.62	***	0.049	*	0.48	***	0.38	*	-0.05	0.27
TAM	CCL2	0.68	***	0.004	0.98	0.2	***	0.34	*	0.3	***	0.08	0.55	-0.04	0.36
	CD68	0.77	***	0.33	*	0.65	***	-0.41	*	0.61	***	0.69	***	0.03	0.59
	IL10	0.49	***	0.36	*	0.008	0.87	-0.13	0.43	0.54	***	0.63	***	-0.03	0.58
M1 Macrophage	INOS (NOS2)	-0.1	0.09	0.03	0.87	-0.02	0.7	-0.23	0.18	0.09	*	-0.2	0.13	0.004	0.93
	IRF5	0.35	***	0.16	0.32	0.28	***	0.092	0.59	0.33	***	0.36	*	0.11	*
	COX2 (PTGS2)	0.17	*	0.19	0.22	0.05	0.37	0.39	*	-0.06	0.19	-0.05	0.71	-0.0	0.47
M2 Macrophage	CD163	0.62	***	0.71	***	0.51	***	0.38	*	0.45	***	0.65	***	-0.07	0.12
	VSIG4	0.8	***	0.58	***	0.52	***	0.16	0.36	0.5	***	0.19	0.14	-0.08	0.09
	MS4A4A	0.84	***	0.7	***	0.73	***	0.21	0.23	0.61	***	0.66	***	-0.07	0.15

Table V. GPNMB related transcription factors(TRRUST)

TF	Target	Model of regulation	Reference(PIMD)
MITF	GPNMB	Activation	18983539
TP53	GPNMB	Unknown	15684612

Figures

Figure 1

GPNMB expression in various cancer types. (A) GPNMB expression in various cancers compared to normal tissues in the Oncomine database. (B) GPNMB expression in different tumors from TIMER (*P < 0.05, **P < 0.01, ***P < 0.001).

Figure 2

Kaplan-Meier survival curves of GPNMB in various cancer types in the Prognoscan (A–H) and Kaplan-Meier plotter databases (I–P). (A–B) Survival curves of OS and RFS in the breast cancer cohort. (C–D) Survival curves of OS and DFS in the colorectal cancer cohort. (E–F) Survival curves of OS and DSS in the lung cancer cohort. (G) OS survival curves of prostate cancer and renal cell carcinoma. (H) OS survival curves of melanoma cancer. (I–J) OS and RFS survival curves of gastric cancer. (K–L) OS and RFS survival curves of LIHC. (M) OS survival curves of breast cancer. (N) OS survival curves of lung adenocarcinoma. (O–P) OS and PFS survival curves of ovarian cancer. OS, overall survival; PFS, progression-free survival; DFS, disease-free survival; RFS, relapse-free survival; DSS, disease-specific survival. DMFS, distant metastasis-free survival; DRFS, distant Recurrence Free Survival.

Figure 3

Association between GPNMB expression and survival status of different cancer types in Prognoscan database.

Figure 4

Multivariable Cox regression analysis in LUAD patients from TCGA.

Figure 5

Association between GPNMB expression and immune infiltration level in BRCA (invasive breast carcinoma), COAD (colon adenocarcinoma), LIHC (hepatocellular carcinoma), LUAD (lung adenocarcinoma), LUSC (lung squamous cell carcinoma), and STAD (stomach adenocarcinoma). (A) GPNMB expression is significantly negatively related to tumor purity and has significant positive correlations with infiltrating levels of B cells, CD8+ T cells, CD4+ T cells, macrophages, neutrophils, and dendritic cells in BRCA. (B) GPNMB expression is significantly negatively related to tumor purity and has significant positive correlations with infiltrating levels of B cells, CD8+ T cells, CD4+ T cells, macrophages, neutrophils, and dendritic cells in COAD. (C) GPNMB expression is significantly negatively related to tumor purity and has significant positive correlations with infiltrating levels of B cells, CD8+ T cells, CD4+ T cells, macrophages, neutrophils, and dendritic cells in LIHC. (D) GPNMB expression is significantly negatively related to tumor purity and has significant positive correlations with infiltrating levels of B cells, CD8+ T cells, CD4+ T cells, macrophages, neutrophils, and dendritic cells in LUAD. (E) GPNMB expression has no significant correlations with tumor purity and infiltrating levels of purity, macrophages, CD4+ T in LUSC. GPNMB expression showed a very weak correlation with B cells, CD8+ T cells, neutrophils, and dendritic cells infiltration level in LUSC. (F) GPNMB expression is significantly negatively related to tumor purity, B cells and has significant positive correlations with infiltrating levels of CD8+ T cells, CD4+ T cells, macrophages, neutrophils, and dendritic cells in STAD.

Figure 6

GPNMB expression is related to macrophage polarization in STAD (stomach adenocarcinoma), LUAD (lung adenocarcinoma) and LUSC (lung squamous cell carcinoma). The immune gene markers: CD86 and CSF1R of monocytes; CCL2, CD68, and IL10 of TAMs; NOS2, IRF5, and PTGS2 of M1 macrophages; CD163, VSIG4, and MS4A4A of M2 macrophages. (A–D) Relationships between GPNMB expression and monocytes (A), TAMs (B), and M1 (C) and M2 macrophages (D) gene markers in STAD. (E–H) Relationships between GPNMB expression and monocytes (E), TAMs (F), and M1 (G) and M2 macrophages (H) gene markers in LUAD. (I–L) Relationships between GPNMB expression and monocytes (I), TAMs (J), and M1 (K) and M2 macrophages (L) gene markers in LUSC. The LUSC acts as the control group, and GPNMB expression is not significantly related to macrophage polarization of LUSC.

Figure 7

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Protein-protein interaction network of GPNMB related transcription factors networks (GeneMANIA). Protein-protein interaction (PPI) network and functional analysis, which indicate the gene enrichment in the target network of GPNMB related transcript factors. Different colors in the network edge signify the bioinformatics methods: physical interactions, co-expression, predicted, co-localization, pathway, genetic interactions and shared protein domains. The different colors of lines signify the enrichment genes biological functions.