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Diagnostic value of vascular endothelial growth factor (VEGF) levels in gastrointestinal cancers with ascites - A cross sectional study

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ABSTRACT

Objective: Malignant ascites is suggestive of peritoneal carcinomatosis. The distinction between malignant and non-malignant ascites in a patient with malignancy is important, as it alters the management and prognosis. Current diagnostic methods are imaging, cytology, and diagnostic laparoscopy, all of which have low sensitivities. The vascular endothelial growth factor (VEGF) is essential for tumour growth and, hence, ascitic VEGF levels can be a diagnostic method for malignant ascites.

Material and Methods: This cross-sectional study was conducted in patients with gastrointestinal malignancies and ascites. The calculated sample size was 68 patients, who were divided into those who were truly positive or negative for malignant ascites based on a composite gold standard, comprising cytology, contrast enhanced computed tomography, and laparoscopy. The ascitic VEGF levels in these patients were compared.

Results: A total of 84 patients were enrolled, of whom 60.71% were found to have malignant ascites. It was found that the greater the volume of ascites, the greater the statistical likelihood of finding truly malignant ascites. The ascitic VEGF levels had a non-normal distribution, with median values of 783.64 and 41.12 pg/mL for malignant and non-malignant ascites ($p<0.001$). Using a receiver operating characteristics curve, a cut-off of 83.68 pg/mL was obtained, with a sensitivity of 100% and a specificity of 93.94%.

Conclusion: This study demonstrates that ascitic VEGF levels are significantly elevated in patients with gastrointestinal malignancies and malignant ascites and hence can reliably be used for diagnosing malignant ascites. This study also shows that massive ascites and well-differentiated tumours have a higher rate of peritoneal carcinomatosis.

Keywords: VEGF, malignant ascites, peritoneal carcinomatosis, biomarkers, gastrointestinal malignancies

INTRODUCTION

Malignant ascites can be defined as malignant cells found in the ascitic fluid, which accounts for approximately 10% of all cases of ascites (1); it suggests peritoneal carcinomatosis. Such patients have a poor prognosis, with a mean survival time after diagnosis of 12-20 weeks (2). However, all patients with malignancies and ascites do not necessarily have malignant ascites as such patients usually have multiple comorbidities and hence have multiple causes for ascites such as hypoalbuminemia, anemia, liver and cardiac dysfunction among others. The distinction between a true malignant ascites and a non-malignant cause of ascites in a patient with malignancy is important to make as it alters the staging of the disease and hence changes the treatment.

Current methods of diagnosing malignant ascites include history and clinical examination, imaging modalities, aspiration cytology and diagnostic laparoscopy (3,4). Among the non-invasive testing methods, cytology has the best specificity, which can go up to 90-100% (5). However, due to its low sensitivity of around 50-60% (6,7), the rates of false-negative results are high, misleading the physician. Clinical symptoms may be atypical, unreliable, and subjective. Current biochemical tests such as albumin, total protein and tumour markers lack sufficient specificity (8). The specificity of computed tomographic (CT) scans for the diagnosis of peritoneal carcinomatosis ranges from 85% to 87%, but its sensitivity lies only around 42% to 68% (9). Therefore, there is a practical challenge of diagnosing malignant ascites with a reliable method.

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Angiogenesis is vital for tumor growth, invasion, and metastasis. Tumor cells produce various angiogenic factors, one of which is the vascular endothelial growth factor (VEGF) (10-12). Review of the literature supports that angiogenesis promoted by VEGF is associated with fluid accumulation in human tumor effusions; malignant ascites is accompanied by high levels of VEGF in these fluids (13). While a few studies have examined VEGF as a biomarker, its diagnostic utility in differentiating malignant from non-malignant ascites in gastrointestinal malignancies remains underexplored (14-16). This study aims to evaluate VEGF levels in ascitic fluid, as a diagnostic tool and compare its utility against the existing methods. Notably, VEGF levels can be measured from the same sample that was collected for cytological assessment, eliminating the need for any additional invasive procedures.

MATERIAL and METHODS

This observational cross-sectional study was conducted in the department of surgery in a tertiary care hospital from March 2020 to December 2021, after obtaining approval from the Institute Ethics Committee (IEC). Written informed consent was obtained from all the patients before the commencement of the study. Patients were given full freedom to withdraw participation at any point. All patients more than 18 years of age, with proven gastrointestinal malignancy and associated ascites, were included. Patients with pre-testing interventions such as radiotherapy, chemotherapy, or surgery and patients with uncontrolled renal, hepatic, and cardiac dysfunction, as well as those with minimal ascites that could not be sampled for analysis, were excluded.

The aim of the study was to evaluate the role of ascitic VEGF levels in patients with gastrointestinal malignancies and concurrent ascites in diagnosing malignant ascites. The primary objective was to determine the sensitivity and specificity of detection of malignant ascites. The secondary objectives were to show the relationship between ascitic fluid VEGF levels and quantity of ascites, as well as between ascitic fluid VEGF levels and differentiation of the primary tumor, and to determine the sensitivity and specificity of malignant cytology and contrast enhanced computed tomography (CECT) in diagnosing malignant ascites.

Participants with proven gastrointestinal malignancy and associated ascites were divided into two outcome groups based on a composite gold standard comprising positive cytology for malignant cells, CT scan findings suggestive of peritoneal carcinomatosis, and diagnostic laparoscopy/laparotomy findings of peritoneal metastasis. Patients testing positive for any of these criteria were classified as having malignant ascites, while those testing negative across all criteria were categorized as non-malignant ascites. Positivity in malignant cytology was defined as the presence of malignant cells in the ascitic fluid. Positivity in a contrast CT scan was defined as the presence of findings suggestive of peritoneal metastasis, pelvic deposits, interbowel deposits, and/or omental deposits caking. Positivity in diagnostic laparoscopy/laparotomy was defined as a visible peritoneal metastasis, pelvic deposits, interbowel deposits, and/or omental deposits or caking, or through cytology of peritoneal washing or proven histopathology of intra-operative peritoneal biopsies.

Patients were selected by convenient sampling from the population that visited the surgery out-patient and emergency services who fit the inclusion criteria. The sample size was calculated to estimate the sensitivity of VEGF to differentiate malignant ascites from non-malignant causes of ascites in patients with gastrointestinal malignancies. Assuming an alpha error of 5% and an expected VEGF sensitivity 91.3%, based on a study done by Dong et al. (15), the minimum required number of diseased subjects was 34 with an absolute precision of 7%. The sample size was calculated using nMaster software version 2.0. Patients continued to be enrolled into the study until a minimum of 34 subjects were present in each outcome group. A total of 84 patients were included in this study.

Demographic and clinical data of the patients were collected. Ascitic fluid was aspirated under sterile conditions for malignant cytology and a part of this sample was taken for analyzing VEGF levels. This sample was immediately centrifuged at 3000 rpm for 15 minutes at 4 °C. Cell-free supernatant was collected, and these aliquots were stored at -40 °C before determination of VEGF levels. The diagnostic biopsies, and staging measures taken by the treating surgeon were also followed up. The kit used was from ELK technologies, an ELISA kit that measured human VEGF-A levels (ELK1129). The working principle of the test is a sandwich enzyme immunoassay. The kit was pre-coated with an antibody specific to VEGF to which standards or samples were added, after which avidin conjugated to horseradish peroxide was added. Those wells that contain VEGF will change in colour from yellow to blue, after which the concentration of VEGF will be determined by spectrophotometric methods.

Statistical Analysis

The median (interquartile range) of the ascitic VEGF levels of both groups was determined and compared using the Mann-Whitney U test to determine whether VEGF levels were elevated in malignant ascites specifically. The sensitivity and specificity of each parameter, i.e., CECT, malignant cytology and VEGF levels were also calculated and compared. All continuous variables, such as age, VEGF levels, etc., were summarized using mean [standard deviation (SD)] or median (interquartile range) depending on the normality of distribution. Categorical variables such as gender, site of tumour, grade, stage of tumour etc. were summarized using proportions (percentage). The mean VEGF

values were compared between the two study groups using Student's t-test and a receiver operating characteristic (ROC) curve was employed to determine appropriate cut-offs of VEGF to label malignant or non-malignant ascites. The sensitivity and specificity of the chosen cut-off was reported along with 95% confidence intervals. The sensitivity and specificity of malignant cytology and CECT for detecting malignant ascites were also calculated by comparing these variables to the composite gold standard and constructing a 2x2 table. All data analysis was done using STATA v.14.

RESULTS

A total of 84 patients were enrolled in the study period; 51 were found to have malignant ascites and 33 had ascites due to other causes, according to the composite gold standard.

Among the 84 participants, 48 (57.14%) were male and 36 (42.86%) were female, showing a slight male preponderance. The mean (± SD) age of the study population was found to be 53.72 (±13.13) years, with 60.71% of patients being less than 60 years. Most patients (62.65%, 52/84) were observed to not have any known medical comorbidities. Among the patients with medical comorbidities, the most common was type 2 diabetes mellitus in 17 patients (20.48%), followed by hypertension in 10 (12.05%) patients (Table 1).

In regard to the organ of origin, the common sites were colon, stomach, and pancreas in that order with 21 (25%), 16 (19.05%), and 12 (14.29%) patients, respectively (Table 2). When the tumour was characterized according to its histopathological types, it was seen that the most common type was adenocarcinoma, with 91.67% (77/84) of tumours falling in this category.

Table 1. Demographic details of the study population

	Truly positive for malignant ascites n (%), T=51	**Truly negative for malignant ascites n (%), T=33**	**Total, n (%) T=84**
Gender			
Male	28 (54.90%)	20 (60.61%)	48 (57.14%)
Female	23 (45.10%)	13 (39.39%)	36 (42.86%)
Age			
Less than 60 years	29 (56.86%)	22 (66.67%)	51 (60.71%)
60 years and above	22 (43.14%)	11 (33.33%)	33 (39.29%)
Co-morbidities			
Nil	32 (62.75%)	20 (60.61%)	52 (62.65%)
Diabetes mellitus	10 (19.61%)	7 (21.21%)	17 (20.48%)
Hypertension	7 (13.73%)	3 (9.09%)	10 (12.05%)
Hepatitis B	1 (1.96%)	1 (3.03%)	2 (2.41%)
Pulmonary tuberculosis	1 (1.96%)	0 (0.00%)	1 (1.2%)

Table 2. Organ of origin of the primary tumour in the study population

Organ of origin	**Truly positive for malignant ascites n (%), T=51**	**Truly negative for malignant ascites n (%), T=33**	**Total, n (%) T=84**
Esophagus	2	1	3 (3.57%)
Stomach	10	6	16 (19.05%)
Colon	9	12	21 (25.00%)
Rectum	3	2	5 (5.95%)
Pancreas	9	3	12 (14.29%)
Liver	2	1	3 (3.57%)
Gastro-esophageal junction carcinoma	4	1	5 (5.95%)
Ampullary carcinoma	2	2	4 (4.94%)
Hilar cholangiocarcinoma	2	1	3 (3.57%)
Distal cholangiocarcinoma	2	0	2 (2.38%)
Intra-hepatic cholangiocarcinoma	2	1	3 (3.57%)
Gall bladder	4	0	4 (4.94%)
Small bowel	0	3	3 (3.57%)

After characterization of their type, the tumours were divided on the basis of their differentiation into well, moderately, or poorly differentiated tumours, as shown in Table 3. Some tumours could not be adequately characterized as belonging to the above groups due to the limited tissue sample obtained by biopsy or fine needle aspiration. The most common group was found in the moderately differentiated tumours, with 26 (40%) patients. However, 19 patients could not be classified into these groups due to the above-mentioned reason.

In all subjects, the ascites was quantified and categorized into three groups radiologically: Minimal, moderate and massive, as shown in Table 4. All three groups had a comparable number of patients. A majority of patients who were negative for malignant ascites had minimal ascites (22/33, 66.67%). The percentage decreased as the quantity of ascites increased, with only one patient having massive ascites, yet this patient remained negative for malignant ascites. This is in contrast to the patients who tested positive for malignant ascites, where a majority of patients had massive ascites (26/51, 50.98%) and the frequency decreased in parallel with the ascites quantity. Six patients had minimal ascites, which was malignant in origin. When applying Fisher's exact test to determine the p-value, this difference in the ascites quantity between the two groups was found to be significant, with a p-value of <0.001.

Using the composite gold standard, out of a total of 84 patients, 51 (60.71%) were found to have malignant ascites while 33 (39.28%) were found to have ascites due to other causes. Each component was also analysed separately. When considering malignant cytology, 58.82% (30/51) of patients with true malignant ascites had positive cytology while a total of 54/84 (64.29%) were found to be negative for malignant cytology. On examining the CECT scans of these patients for signs of peritoneal carcinomatosis such as omental nodules and peritoneal deposits, a total of 32 patients out of the 51 had true malignant ascites (62.74%) were found to have signs suggestive of peritoneal carcinomatosis, while a total of 19 patients (37.26%) did not have such findings in their scans. If a patient tested positive in either of the categories above, they were not considered for diagnostic laparoscopy/laparotomy, with 38/84 (45.24%) patients fulfilling this criterion. Out of the remaining 46 subjects, 13 patients (28.26%) were found to have peritoneal disease intraoperatively while the remaining 33 (71.74%) did not have the same (Table 5). The sensitivity of malignant cytology and CECT was found to be 58.82% and 62.74%, respectively, while both methods were found to have 100% specificity.

The VEGF levels were found to have a non-normal distribution, and hence, the median and interquartile range were calculated for both the truly positive and negative groups, which were found to be 783.64 (655.94-875.64) pg/mL and 41.12 (35.33-46.12) pg/mL, respectively. These values were found to be significantly different between the two groups, with a p-value of <0.001, calculated using the Mann-Whitney U test. Using the values obtained, a ROC curve was plotted (Figure 1). It was seen that at a cut-off value of 83.68 pg/mL, the area under the curve was 0.9964 with a standard error of 0.0032, sensitivity of 100%, and a specificity of 93.94%.

DISCUSSION

In our study, out of 84 patients, 51 (60.71%) were positive for malignant ascites. This indicates that when patients with gastrointestinal cancers are found to have ascites, the possibility of peritoneal carcinomatosis must be seriously considered, which is consistent with the study conducted by Zhang et al. (6) where they found that 66.7% of patients with malignancy-related ascites had peritoneal carcinomatosis. When analysing

Table 3. Differentiation of the primary tumour in the study population

Differentiation of primary tumour	Truly positive for malignant ascites n (%), T=51	Truly negative for malignant ascites n (%), T=33	Total, n (%) T=84
Well-differentiated	14 (27.45%)	9 (27.27%)	23 (35.38%)
Moderately differentiated	13 (25.49%)	13 (39.39%)	26 (40%)
Poorly differentiated	4 (7.84%)	12 (36.36%)	16 (24.62%)
Not known	2 (3.92%)	17 (51.51%)	19 (22.62%)

Table 4. Quantification of ascites in the study population

Ascites quantification	Truly positive for malignant ascites n (%), T=51	Truly negative for malignant ascites n (%), T=33	Total, n (%) T=84	p-value (Fischer's exact)
Minimal	6 (21.43%)	22 (78.57%)	28 (33.33%)	<0.001
Moderate	19 (65.52%)	10 (34.48%)	29 (34.52%)	
Severe	26 (96.30%)	1 (3.70%)	27 (32.14%)	

the separate components, 35.71% of patients were found to have positive cytology, bringing the sensitivity up to 58.82% with a specificity of 100%. Regarding CECT findings for diagnosis of malignant ascites, we found a sensitivity of 62.74% and a specificity of 100%. This may be due to the subjectiveness of the reporting of CT images, which varies between radiologists, according to their level of expertise. Both of these results are in accordance with published literature (17,18). Diagnostic laparoscopy or laparotomy has a higher diagnostic accuracy (82.2% to 96.6%) (19) and a good sensitivity of up to 92% but involves a measure of operative risk and morbidity to patients who have a limited lifespan. Hence, this method is not generally used as a first-line investigation for patients with gastrointestinal malignancies and ascites but rather as a last resort in patients who have multiple causes of ascites (20). As has been demonstrated, no one method has an adequate sensitivity to use as a diagnostic gold standard, which is also non-invasive and simple to apply. Therefore, we chose to use a composite gold standard to improve the overall sensitivity of the tests as a benchmark against which we compared our test, namely ascitic VEGF levels.

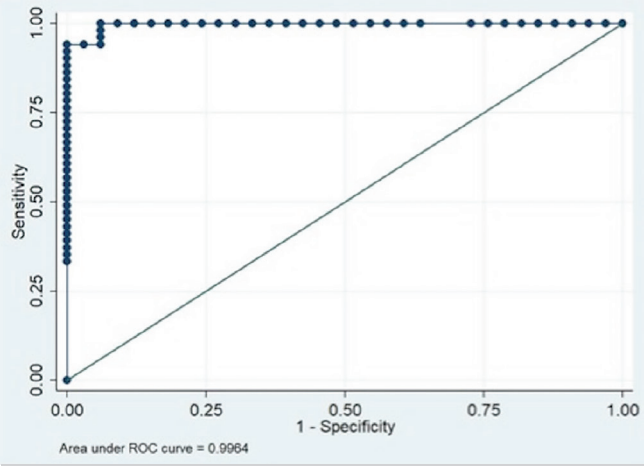

Figure 1. Receiver operating characteristic curve of ascitic VEGF levels in the study population.

VEGF: Vascular endothelial growth factor, ROC: Receiver operating characteristic

Table 5. Results of the composite gold standard in the study population

	n (%), T=84
Malignant cytology	
Positive	30 (35.71%)
Negative	54 (64.29%)
CECT report for malignant ascites	
Positive	32 (38.10%)
Negative	52 (61.90%)
Intra-operative findings suggestive of peritoneal metastasis	
Positive	13 (15.48%)
Negative	33 (39.29%)
Not applicable	38 (45.24%)
Composite gold standard results	
Truly positive	51 (60.71%)
Truly negative	33 (39.29%)

CECT: Contrast enhanced computed tomography

The VEGF is a dimeric, angiogenic glycoprotein with an average molecular mass of around 40,000 kD, which has been found to have stimulatory effects on neovascularization, capillary formation, as well as mitogenic and chemotactic effects on the endothelial cells of blood vessels. All of these actions lead to an increase in the permeability of these cells (21,22). It has been seen that an overexpression of VEGF in tumour cells allows the tumour to meet the high oxygen demands of its growth. As it increases permeability, it also forms an important part of the pathophysiology of malignant ascites, leading to speculation about its differential levels in malignant and non-malignant ascites (23). This is the issue we aimed to address in this study. When ascitic VEGF levels were compared between the patients with true malignant ascites and ascites due to other causes, the median and interquartile range values were found to be 783.64 (655.94-875.64) and 41.12 (35.33-46.12), respectively, and these differences were statistically significant. Using the values obtained, a ROC curve was plotted. It was seen that at a cut-off value of 83.68 pg/mL, the area under the curve was 0.9964 with a standard error of 0.0032, sensitivity of 100% and a specificity of 93.94%. There are just a handful of published studies that correlate ascitic fluid VEGF levels to the occurrence of malignant ascites, all of which compare ascitic VEGF levels between patients with benign and malignant pathologies, in contrast to our study, which exclusively includes patients with gastrointestinal malignancies (14,15,23-26).

We had classified tumours according to their level of differentiation. Out of 23 patients with well-differentiated tumours, 14 (60.87%) were found to be truly positive for malignant ascites, while an equal number of patients with moderately differentiated tumours were found to have both truly malignant and non-malignant ascites. Additionally, a majority of patients with poorly differentiated tumours [12/16 (75%)] were truly negative for malignant ascites. This association was found to be statistically significant. This analysis of the relationship between the differentiations of primary tumour and the presence of peritoneal carcinomatosis has not yet been conclusively proved in available literature, making this a novel finding in this study. However, three types of peritoneal spread of tumours have been elucidated based on tumour grade, namely: Random proximal distribution, seen in moderate and high-grade cancers; complete redistribution in well-differentiated tumours; and widespread cancer distribution that usually occurs in mucinous tumours. The first type is when cancer cells adhere

to the peritoneum near the local area; the second is where there is no adhesion with the peritoneum in the local area due to low metabolic activity of the tumour, leading to more widespread peritoneal dissemination rather than local disease. The last type -widespread cancer distribution- is in which the presence of adherence markers along mucus production leads to the widespread and aggressive dissemination of the tumour (19). This matches our results, with well-differentiated tumours showing a higher proportion of malignant ascites due to peritoneal carcinomatosis.

The ascites were quantified in all subject patients using radiological methods and divided into minimal, moderate, and massive. the patients were almost evenly distributed among the three groups. However, within these three groups, it was seen that a majority of patients (78.57%) with minimal ascites were found to be negative for malignant ascites while 96.3% of patients with massive ascites were found to have true malignant ascites. In other words, as the quantity of ascites increases, the higher the probability is that the patient has peritoneal carcinomatosis. This association was found to be statistically significant by Fischer's exact test. This relationship has not been studied in the literature previously and is therefore a novel finding of this study.

Peritoneal metastases from gastrointestinal cancers are often associated with malignant ascites due to VEGF-related angiogenesis and enhanced vascular permeability. The role of heated intraperitoneal chemotherapy (HIPEC), administered along with cytoreductive surgery, in patients with malignant ascites has been evaluated in many recent studies. HIPEC typically involves the circulation of heated chemotherapeutic agents at temperatures between 41-43 °C within the peritoneal cavity to enhance drug penetration, disrupt VEGF-mediated pathways, and improve local tumor control (27). The heat increases the cytotoxic effects of chemotherapy by improving drug absorption, impairing DNA repair in cancer cells, and reducing peritoneal tumor burden. Another advantage of HIPEC is that this technique reduces the systemic side effects of toxic chemotherapy as the drugs are instilled locally into the peritoneal cavity. HIPEC has shown benefits in select patients, especially with ovarian and appendiceal cancers, but its effectiveness in colorectal cancer remains debated, as seen in the PRODIGE 7 trial, and its role in pancreatic cancer is unclear (28,29).

Pressurized intraperitoneal aerosolized chemotherapy has emerged as a minimally invasive alternative to HIPEC, offering improved drug distribution and deeper tissue penetration, with potential synergy when combined with anti-VEGF therapies such as bevacizumab (30). Other therapeutic options in patients with malignant ascites include systemic chemotherapy, immunotherapy, peritoneo-venous shunting, and diuretics; but each has variable success and risks, necessitating further research to refine treatment protocols and personalize therapy based on tumor biology and patient response (31).

The strengths of our study were that we had a relatively large sample size with a total of 84 patients. They were also a heterogeneous group, with all patients having gastrointestinal malignancies, hence avoiding confounding factors and leading to more reliable results. We had a well-defined composite gold standard to compare ascitic VEGF levels, composed of three checkpoints to have dependable results.

Study Limitations

The limitations of our study were that we did not draw any correlation between the post-diagnosis survival time and VEGF levels. Another area that we did not study was the correlation between serum and ascitic VEGF levels, as well as the effect of the interventions, such as hyper-thermic intraperitoneal chemotherapy, including chemotherapy on the ascitic VEGF levels. An area in which we would like to conduct further research is the potential therapeutic value of using anti-VEGF agents in the palliation of malignant ascites.

CONCLUSION

In this study, we demonstrated that ascitic VEGF levels are significantly elevated in patients with gastrointestinal malignancies and malignant ascites. With a sensitivity of 100% and specificity of 93.94%, ascitic VEGF proved to be a highly reliable biomarker for diagnosing malignant ascites. These findings suggest that VEGF can enhance early diagnosis and potentially open avenues for targeted therapeutic interventions in managing malignant ascites.

Ethics

Ethics Committee Approval: This observational cross-sectional study was conducted in the department of surgery in a tertiary care hospital from March 2020 to December 2021, after obtaining approval from the Institute Ethics Committee (IEC).

Informed Consent: Written informed consent was obtained from all the patients before the commencement of the study.

Footnotes

Author Contributions

Concept - S.S.; Design - V.P.N.; Supervision - V.P.N.; Materials - N.H.; Data Collection or Processing - E.M.K.S.; Analysis or Interpretation - S.S.; Literature Search - E.M.K.S.; Critical Review - V.P.N.; Writing - E.M.K.S.

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REFERENCES

1. *Stukan M. Drainage of malignant ascites: patient selection and perspectives. Cancer Manag Res. 2017;9:115-130.*
2. *Sangisetty SL, Miner TJ. Malignant ascites: A review of prognostic factors, pathophysiology and therapeutic measures. World J Gastrointest Surg. 2012;4:87-95.*

3. *European Association for the Study of the Liver. EASL clinical practice guidelines on the management of ascites, spontaneous bacterial peritonitis, and hepatorenal syndrome in cirrhosis. J Hepatol. 2010;53:397-417.*
4. *Moore KP, Aithal GP. Guidelines on the management of ascites in cirrhosis. Gut. 2006;55(Suppl 6):vi1-vi12.*
5. *Markal AC, Dideban B, Memdani A, Rathi K, Baskaran N A rare case of malignancy-associated ascites with no identifiable cause in a 29-year-old female with BRCA1 mutation. J Case Rep Images Oncology. 2023;9:27-31.*
6. *Zhang F, Feng Z, Zhang Y, Liu Z, Sun X, Jin S. Determination of the optimal volume of ascitic fluid for the precise diagnosis of malignant ascites. Saudi J Gastroenterol. 2019;25:327-332.*
7. *Rooper LM, Ali SZ, Olson MT. A specimen volume of ≥80 mL improves cytologic sensitivity for malignant ascites: a retrospective analysis of 2665 cases. J Am Soc Cytopathol. 2016;5:301-305.*
8. *Jacob R, A S, Abdul Razack N, Prabhuswamimath SC. Malignancy of malignant ascites: A comprehensive review of interplay between biochemical variables, tumor microenvironment and growth factors. Asian Pac J Cancer Prev. 2024;25:3413-3420.*
9. *Veron Sanchez A, Bennouna I, Coquelet N, Cabo Bolado J, Pinilla Fernandez I, Mullor Delgado LA, et al. Unravelling peritoneal carcinomatosis using cross-sectional imaging modalities. Diagnostics (Basel). 2023;13:2253.*
10. *Niu G, Chen X. Vascular endothelial growth factor as an anti-angiogenic target for cancer therapy. Curr Drug Targets. 2010;11:1000-1017.*
11. *Ghalehbandi S, Yuzugulen J, Pranjol MZI, Pourgholami MH. The role of VEGF in cancer-induced angiogenesis and research progress of drugs targeting VEGF. Eur J Pharmacol. 2023;949:175586.*
12. *Liu ZL, Chen HH, Zheng LL, Sun LP, Shi L. Angiogenic signaling pathways and anti-angiogenic therapy for cancer. Signal Transduct Target Ther. 2023;8:198.*
13. *Sherer DM, Eliakim R, Abulafia O. The role of angiogenesis in the accumulation of peritoneal fluid in benign conditions and the development of malignant ascites in the female. Gynecol Obstet Invest. 2000;50:217-224.*
14. *Zhan N, Dong WG, Wang J. The clinical significance of vascular endothelial growth factor in malignant ascites. Tumour Biol. 2016;37:3719-3725.*
15. *Dong WG, Sun XM, Yu BP, Luo HS, Yu JP. Role of VEGF and CD44v6 in differentiating benign from malignant ascites. World J Gastroenterol. 2003;9:2596-2600.*
16. *Guo YY, Peng XL, Zhan N, Tian S, Li J, Dong WG. Development and validation a simple model for identify malignant ascites. Int J Med Sci. 2021;18:1966-1974.*
17. *Diop AD, Fontarensky M, Montoriol PF, Da Ines D. CT imaging of peritoneal carcinomatosis and its mimics. Diagn Interv Imaging. 2014;95:861-872.*
18. *Reginelli A, Giacobbe G, Del Canto MT, Alessandrella M, Balestrucci G, Urraro F, et al. Peritoneal carcinosis: What the radiologist needs to know. Diagnostics (Basel). 2023;13:1974.*
19. *Desai JP, Moustarah F. Peritoneal Metastasis. In: StatPearls. [Internet]. Treasure Island (FL): StatPearls Publishing; [cited 8 Feb 2022]. Available from: http://www.ncbi.nlm.nih.gov/books/ NBK541114/*
20. *Abdelkader AM, Zaher NA, Mitwally RA, Hassanien SEA, Dawa SK. The role of laparoscopy in the diagnosis of ascites of unknown etiology. The Egyptian Journal of Surgery. 2019;38:760-765.*
21. *Wang X, Bove AM, Simone G, Ma B. Molecular bases of VEGFR-2-mediated physiological function and pathological role. Front Cell Dev Biol. 2020;8:599281.*
22. *Gamde SM, Ogenyi OD. Angiogenesis in breast cancer: A review. Asian Pacific Journal of Cancer Biology; 2024;9:97-103.*
23. *Bekes I, Friedl TW, Köhler T, Möbus V, Janni W, Wöckel A, et al. Does VEGF facilitate local tumor growth and spread into the abdominal cavity by suppressing endothelial cell adhesion, thus increasing vascular peritoneal permeability followed by ascites production in ovarian cancer? Mol Cancer. 2016;15:13.*
24. *Karatolios K, Pankuweit S, Moosdorf RG, Maisch B. Vascular endothelial growth factor in malignant and benign pericardial effusion. Clin Cardiol. 2012;35:377-381.*
25. *Nascimento I, Schaer R, Lemaire D, Freire S, Paule B, Carvalho S, et al. Vascular endothelial growth factor (VEGF) levels as a tool to discriminate between malignant and nonmalignant ascites. APMIS. 2004;112:585-587.*
26. *Lee HK, Chae HS, Kim JS, Kim HK, Cho YS, Rho SY, et al. Vascular endothelial growth factor levels in ascites between chemonaive and chemotreated patients. Yonsei Med J. 2008;49:429-435.*
27. *Aziz MB, Di Napoli R. Hyperthermic intraperitoneal chemotherapy. StatPearls. StatPearls: Publishing; 2023.*
28. *Quénet F, Elias D, Roca L, Goéré D, Ghouti L, Pocard M, et al. Cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy versus cytoreductive surgery alone for colorectal peritoneal metastases (PRODIGE 7): a multicentre, randomised, open-label, phase 3 trial. Lancet Oncol. 2021;22:256-266.*
29. *Sikora A, Sullivan KM, Dineen S, Raoof M, Karolak A. Emerging therapeutic approaches for peritoneal metastases from gastrointestinal cancers. Mol Ther Oncol. 2024;32:200767.*
30. *Daniel SK, Sun BJ, Lee B. PIPAC for gastrointestinal malignancies. J Clin Med. 2023;12:6799.*
31. *Berger JM, Preusser M, Berghoff AS, Bergen ES. Malignant ascites: Current therapy options and treatment prospects. Cancer Treat Rev. 2023;121:102646.*