

Comparison of Patients with Ultrasound Guided Tru-Cut Biopsy / Acid Cytology with Final Pathology Results from Patients Operated with Suspicious for Ovarian Cancer

Fatma Basak Tanoglu ^{1*}, Gurkan Kiran ¹, Caglar Cetin ¹, Burcu Gul ², Ozge Pasin ³, Temel Fatih Yilmaz ⁴

¹ Department of Obstetrics and Gynecology, Bezmialem University Faculty of Medicine, İstanbul, Turkey

² Department of Pathology, Bezmialem University Faculty of Medicine, İstanbul, Turkey

³ Department of Biostatistics, Bezmialem University Faculty of Medicine, İstanbul, Turkey

⁴ Department of Interventional Radiology, Bezmialem University Faculty of Medicine, İstanbul, Turkey

***Corresponding Author:** Fatma Basak Tanoglu, M.D., Bezmialem Vakif University Hospital İskender Paşa Mh Adnan Menderes Bulvarı, Vatan Cad, 34093 Fatih, İstanbul, 34093, Turkey.

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Abstract

Objective

In women who are believed to have ovarian cancer but have poor performance status or have advanced disease believed to be beyond the scope of primary cytoreductive surgery, NACT can be given to patients with acid cytology and/or tru-cut biopsy referral. Our aim is to determine the accuracy, adequacy, safety and reliability of these minimally invasive interventional procedures.

Material and Methods

This is a retrospective analysis of 63 patients with a suspicious of ovarian cancer in our hospital between 2014 and 2021, who underwent ultrasound-guided acid cytology and tru-cut biopsy, and also had postoperative final pathology results.

Results

Comparing acid cytology, tru-cut biopsy, acid cytology and tru-cut biopsy at the same time with the postoperative final pathology results, it was seen that the PPV was 100% in all groups. It was revealed that the sensitivity of acid cytology was 64%, the specificity was 100%, the NPV was 12%, and the accuracy of the test was 65%. The sensitivity of the tru-cut biopsy was 91%, the specificity was 100%, the NPV was 42%, and the accuracy of the test was 92%. In the case of both procedures, the sensitivity was calculated as 93% and the accuracy of the test was calculated as 93%. There were no false positive cytology and biopsy results that could lead to unnecessary NACT therapy in the study.

Conclusion

Minimally invasive procedures can be safely applied to patients with low adverse event and high accuracy rates, since they provide NACT in patients who are thought to be candidates for interval surgery.

Keywords: ovarian cancer, tru-cut biopsy, acid cytology

Introduction

Ovarian cancer ranks 4th among the deadliest cancers in women and has the highest mortality rate of all gynecological malignancies. Patients do not admit to the hospital at an early stage due to their nonspecific symptoms, so only 30% of cases can be diagnosed at stage I or II, and the majority of ovarian cancer cases are diagnosed at an advanced stage [1].

It is a disease with a poor prognosis since it can be diagnosed at an advanced stage, with high recurrence rates despite treatment, low disease-free and overall survival rates in the advanced stage. The primary treatment is primary debulking surgery or interval debulking surgery after neoadjuvant chemotherapy, depending on the stage of the tumor, optimal

cytoreduction status, and comorbidities of the patients. Today, image-guided biopsy/ascites cytology application using ultrasound (USG) or computed tomography (CT) guidance is very valuable in the presence of an advanced-stage ovarian cancer suspicion. In most cases, histological diagnosis can be achieved without diagnostic laparoscopy or laparotomy by providing a site-specific primary tumor diagnosis. Tissue sampling by diagnostic laparoscopy or laparotomy requires general anesthesia and hospitalization, resulting in higher costs and potential surgical morbidity. Also, diagnostic laparoscopy in advanced ovarian cancer is associated with port-site metastases in up to 17-49% of the patients [2,3].

The present study was designed to compare the tru-cut biopsy/ascites cytology samples performed with preoperative ultrasound guidance and the postoperative final pathology results of the patients operated on with a preliminary diagnosis of ovarian cancer. The primary aim was to compare the consistency of tru-cut biopsy and ascites cytology results with the final pathology results. Our secondary aim were determining post-procedure bleeding, blood transfusion, hospitalization requirement, re-operation, infection, sepsis or death (safety); the power of pathology samples taken by interventional procedures to indicate biology, origin, and tumor type (reliability); the consistency of the reports of acid cytology and tru-cut biopsy materials with the final postoperative pathologies (accuracy); whether sufficient cells and tissues were obtained so that the materials taken could be examined by pathologists (proficiency).

Material and Methods

This study was a retrospective analysis of patients with suspicious ovarian cancer who had a tru-cut biopsy, ascites cytology results, and postoperative final pathology results in the Department of Obstetrics and Gynecology in our hospital between January 2014 and December 2021. The Non-interventional Ethics Committee decision number of the study was 2022/31. Data were collected by reviewing electronic patient records, including USG/MR/CT/PET reports, laboratory tests, pathology/surgery reports, and hospitalizations in other parts of our hospital. Written informed consent was obtained from all patients before the procedure.

The study included patients aged 18-85 years, those who were evaluated primarily in favor of ovarian cancer with imaging, laboratory tests, and physical examination findings but considered not possible to achieve optimal cytoreduction, those with a previous history of malignancy and suspected recurrence, those who were not suitable for primary debulking surgery due to comorbidities and performance status, and those who underwent tru-cut biopsy and/or acid cytology to make the differential diagnosis of benign and malignant diseases that were clinically and radiologically similar to ovarian cancer. These patients were also patients with final pathology who had been operated on with or without neoadjuvant chemotherapy (NACT). Patients who were considered to have ovarian cancer based on imaging, laboratory tests, and physical examination findings but underwent staging surgery or primary debulking surgery, those who had tru-cut and ascites cytology results but followed up with NACT without surgery, and those who were followed up closely with routine controls without additional treatment or lost to follow-up were excluded.

The patients' demographic characteristics (age, body mass index, comorbidity, performance status of patients-ECOG score, history of malignancy), physical examination findings, laboratory test results, and imaging findings (complaint, presence of ascites, CA125 values, clinical-stage), the site of the biopsy obtained, histopathological features in the tru-cut biopsy, ascites cytology, and final pathology reports, operation-related variables (degree of cytoreduction, adverse event, blood transfusion, ICU need, hospitalization time) were analyzed through the hospital database. The statistical evaluation was performed accordingly.

Ascites Cytology Technique

Samples were obtained by paracentesis from patients with diffuse ascites in the abdomen detected with imaging studies. In the Interventional Radiology Department of our hospital, following the determination of the localization where the procedure would be performed by abdominal USG, immediately after local anesthesia was applied to this area with Locanest Spray containing 10% Lidocaine, paracentesis guided by abdominal USG was performed with a 20 Gauge Spinal Needle Quincke 6 Inc to collect at least 60cc of ascites fluid.

Tru-Cut Biopsy Technique

Abdominal USG-guided tru-cut biopsy was taken from patients with omental cake, peritoneal carcinomatosis-peritoneal implant, pelvic mass, liver implant, or pathological lymph node, which were determined to be accessible by USG in imaging studies. INR (International Normalized Ratio) and platelet values of the patients were routinely checked before the procedure. The biopsy was not performed if INR>1.5 or platelet<50,000 cells/mL in the peripheral blood of the patients. In patients using anticoagulants, the drug was discontinued five days before and restarted after the procedure. In the Interventional Radiology Department of our hospital, following the determination of the localization where the procedure would be performed with abdominal USG, 5 minutes after local anesthesia was applied to this area with 3cc subcutaneous injection of 2% Prilocaine Hydrochloride, a tru-cut biopsy guided by abdominal USG was performed with the help of 17G TruGuide Bard coaxial needle and 18G Max-Core gun with at least two tissue cylinders 20mm long were obtained (**Figure 1**).

In the study, radiologists who performed USG-guided paracentesis and tru-cut biopsy procedures have 10, 20 and 25 years of experience respectively. As indicated in previous studies, we know that performing these procedures by experienced radiologists, especially under USG guidance, has a crucial role in tissue sufficiency [4].

Histopathological Evaluation

The procedure of Cytological Preparations: After the incoming cytological material was identified macroscopically, it was centrifuged at 2500 rpm for 10 minutes. The supernatant on it was spilled. The remaining cells were mixed, and one drop was taken and spread on two slides. The slides were quickly put into 96% alcohol. The slides were kept for one night and stained in the 'PAP Procedure' of the automatic stainer the following day. Immediately after the smear, 8 ml of rejection solution was placed in the patient sample in the tube, then 15 drops of citrated plasma were dropped. Then, the 'formol-alcohol' mixture prepared before was added to this mixture to fill it. After being centrifuged at 600 rpm for 10 minutes and left for at least 5 hours, the tissue was taken as 'Cell Block' for follow-up. While cytological smears were stained with the PAP staining technique, cell blocks were stained using the Hematoxylin-Eosin technique.

Hematoxylin Eosin procedure: The recommended tissue fixative for paraffin-embedded blocks was 10% neutral buffered formalin (NBF). Sections of 2 µm were taken on slides from the blocks fixed and embedded with 10% NBF. Slides were stained in the automatic stainer and sealed in the sealing device.

For immunohistochemistry application, 2-micron sections were taken on positively charged slides. Necessary immunohistochemical analyzes were performed on these sections using DAB chromogen on Ventana Benchmark XT and Ventana Benchmark Ultra devices (**Figure 2**).

All cytology and tissue samples obtained in our study were examined by two experienced pathologists. All tru-cut biopsy and acid cytology materials examined by the pathologist; reported detailing tumor origin and histological subtype.

Statistical Evaluation

The descriptive statistics of the variables in the qualitative structure in the study were presented as numbers and percentages, and the descriptive statistics of the quantitative structure as mean, median, standard deviation, minimum and maximum values. The conformity of the quantitative variables to the normal distribution was examined using the Shapiro Wilk test. The Mann-Whitney U test compared the groups consisting of two categories in terms of the relevant quantitative variable means. Pearson chi-square and Fisher exact chi-square tests were used for comparisons between groups regarding the incidence of subgroups of related variables. Sensitivity, specificity, accuracy, positive predictive value, and negative predictive values were calculated with confidence intervals for evaluating ascites cytology and tru-cut biopsy techniques in terms of final pathology result. The statistical significance level was considered 0.05, and the SPSS (version 28) package program was used in the analyses.

Results

Between January 2014 and December 2021 a total of 76 patients were performed at our department. Sixty-three patients were included in the study after exclusion (**Figure 3**). The patients were divided into 3 groups. In 12 of these, only acid cytology was obtained before the operation. Only tru-cut biopsy was taken in 22 of them. In 29, both acid cytology and tru-cut biopsy were taken. As a result, paracentesis was performed in 41 patients and trucut biopsy was performed in 51 patients.

The mean age of the 63 female patients included in the study was 62 years, the mean body mass index was 28.5 kg/m², and the mean serum CA125 value was 1546 U/ml. (**Table 1**). The complaints, comorbidities, presence of preoperative ascites, ECOG scores, clinical stages, and biopsy sites of the patients are presented in (**Table 2**). Tru-cut biopsy was obtained from the omentum in 15 patients, peritoneum in 24, pelvic mass in 8, the liver in 2, right cervical LAP (lymphadenopathy) in 1, and left inguinal LAP in 1 patient.

In the evaluation, the ascites cytology result of 23 of 30 patients whose final pathology result was reported as 'serous type ovarian cancer' was 'malignant.' When the patients whose histopathological type was reported as 'serous' were compared with the others, the rate of 'malignant' ascites cytology results was statistically significantly higher. (p=0.001) The tru-cut biopsy results of 39 of 42 patients whose final pathology result was reported as 'serous type ovarian cancer' were 'malignant.' When the patients whose histopathological type was reported as 'serous' were compared with the others, the rate of 'malignant' tru-cut biopsy results was also statistically significant (p=0.013)(**Table 3**).

Among the patients included in the study, the final pathology results of 5 patients were reported as benign. These patients were thought to have ovarian cancer clinically and radiologically.

Ascites cytology was obtained in 41 of the 63 patients included in the study. In 25 of these patients, both the ascites cytology and final pathology

results were reported as 'malignant' (true positive cytology). In two patients, both ascites cytology and final pathology results were reported as 'benign' (true negative cytology). Incompatibility was present in 14 patients. The final pathology results of these patients whose ascites cytology was 'nonmalignant' were reported as 'malignant' (false negative cytology). There was no patient whose ascites cytology result was 'malignant' but whose final result was reported as 'nonmalignant' (false-positive cytology). Tru-cut biopsy was taken in 51 of the patients. In 44 of 51 patients, both tru-cut biopsy and final pathology results were reported as 'malignant' (true positive biopsy). In three patients, both tru-cut biopsy and final pathology results were reported as 'nonmalignant' (true negative biopsy). There was incompatibility in 4 patients. The final pathology results of these patients whose tru-cut biopsy results were 'nonmalignant' were reported as 'malignant' (false-negative biopsy). There was no false-positive biopsy. Both ascites cytology and tru-cut biopsy were taken in 29 of the patients. In the evaluation where the result was considered as 'malignant' if at least one of the two procedures was 'malignant,' while malignancy consistent with the final pathology results was observed in 27 patients (true positive), ascites cytology and tru-cut biopsy were found to be 'nonmalignant' in 2 patients, and the final pathology result was reported as 'malignant' (false negative). True negativity could not be evaluated since no patients in this group had a 'nonmalignant' final result. False positivity was not present. In this group, both acid cytology and tru-cut biopsy results of 17 patients were reported as malignant. Acid cytology and tru-cut biopsy results of 2 patients were benign. In 8 patients whose tru-cut result was malignant, acid cytology result was benign, and in 2 patients whose tru-cut result was benign, acid cytology result was malignant. No statistically significant agreement was observed between acid cytology and tru-cut. (p=0.482; kappa=0.11).

When the pathology results of patients with acid cytology, tru-cut biopsy, acid cytology and tru-cut biopsy were compared with the postoperative final pathology results, we found that the PPV was 100% in all three groups. We found that the sensitivity of the acid cytology was 64%, the specificity was 100%, the NPV was 12% and the accuracy of the test was 65%. We found that the sensitivity of the Tru-cut biopsy was 91%, the specificity was 100%, the NPV was 42%, and the accuracy of the test was 92%. In the case of both procedures, we found that the sensitivity was 93% and the accuracy of the test was 93%. However, specificity and NPV could not be calculated since there were no patients in the third group whose final pathology result was reported as benign. (**Table 4**).

When the final pathology result was 'malignant,' the optimal rate for surgical cytoreduction was evaluated to be significantly higher in the patients who received NACT than those who did not (p=0.007). Also, the rate of postoperative hospitalization in less than ten days was significantly higher in the NACT group (p=0.001). However, no statistically significant difference was observed between the two groups in terms of perioperative/postoperative adverse events, intraoperative/postoperative blood transfusion, and ICU needs (**Table 5**).

Parameters (n=63)	Mean Value \pm SD**	Range
Age (years)	62.5 \pm 11.2	32 - 81
BMI (kg/m ²)	28.6 \pm 5.1	16.2 - 44.1
CA125 (U/ml)	1546.0 \pm 2365.0	7.2 - 12230.0

*Body Mass Index

** Standard deviation

Table 1. Age, BMI, and CA125 values of the patients.

Parameters		n (%)
Complaint	Pain, swelling, palpable mass	42 (66.7)
	Constipation, diarrhea, weight loss	3 (4.8)
	Urinary complaint, abnormal uterine bleeding	6 (9.5)
	Loss of appetite	7 (11.1)
	Shortness of breath	5 (7.9)
Comorbidity*	No	25 (39.7)
	Yes	38 (60.3)
Preoperative Ascites	No	7 (11.1)
	Yes	56 (88.9)
ECOG** Score	0	10 (15.9)
	1	27 (42.9)
	2	21 (33.3)
	3	5 (7.9)
Clinical Stage	2	8 (12.7)
	3	38 (60.3)
	4	17 (27)
Biopsy Location	Omentum	15 (23.9)
	Peritoneum	24 (38.1)
	Mass	8 (12.7)
	Other	4 (6.3)

*Comorbidity defined as hypertension, diabetes mellitus, cardiac, metabolic and cerebrovascular disorders.

**Eastern Cooperative Oncology Group

Table 2. Preoperative evaluation of patients and biopsied sites.

Histopathology n (%)	Ascites Cytology (n=41)		p	Tru-Cut Biopsy (n=51)		p
	Non-malignant (n=16)	Malignant (n=25)		Non-malignant (n=7)	Malignant (n=44)	
Serous type	7 (43.8)	23 (92.0)	0.001	3 (42.9)	39 (88.6)	0.013
Others	9 (56.3)	2 (8.0)		4 (57.1)	5 (11.4)	

p ≤ 0.05 significant difference, comparison of groups.

Table 3. Comparison of ascites cytology and tru-cut biopsy association with serous ovarian cancer and other types.

Procedures	Sensitivity	Specificity	PPV	NPV	Accuracy
	(95% CI) (%)	(95% CI) (%)	(95% CI) (%)	(95% CI) (%)	(%)
Ascites cytology (n=41)	64.1 (47.1-78.3)	100 (19.7-100)	100 (83.4-100)	12.5 (2.2-39.5)	65.9
Tru-cut biopsy (n=51)	91.7 (79.1-97.2)	100 (30.9-100)	100 (89.9-100)	42.8 (11.8-79.7)	92.2
Ascites cytology + tru-cut biopsy (n=29)	93.1 (75.7-98.7)	N/A	100 (84.4-100)	N/A	93.1

*Positive predictive value

**Negative predictive value

Patients who underwent both procedures were included in the first and second groups.

Table 4. Evaluation of sensitivity, specificity, PPV, NPV, and accuracy of ascites cytology and tru-cut biopsy.

PARAMETERS n(%)	NACT + (n=41)	NACT – (n=22)	p-value
Cytoreduction			0.007
• Optimal	11 (64.7)	38 (92.7)	
• Suboptimal	6 (35.3)	3 (7.3)	
Perioperative Complication	6 (35.3)	12 (29.3)	0.652
Postoperative Complication	6 (35.3)	10 (24.4)	0.398

Intraoperative Transfusion	5 (29.4)	4 (9.8)	0.060
Postoperative Transfusion	8 (47.1)	12 (29.3)	0.194
ICU** Need	11 (64.7)	21 (51.2)	0.347
Prolonged Hospitalization			0.001
• Under 10 days	7 (41.2)	34 (82.9)	
• Over 10 days	10 (58.8)	7 (17.1)	

*Neoadjuvant chemotherapy

**Intensive Care Unit

Since the final histopathology results of 5 patients in the group that did not receive neoadjuvant chemotherapy were reported as benign, they were excluded from the table.

p ≤ 0.05 significant difference, comparison of groups.

Table 5. Postoperative evaluation in patients with and without NACT.

Discussion

In recent years, interval debulking surgery has become an option for advanced ovarian cancer. Randomized controlled studies in large populations have revealed that it provides advantages in providing optimal cytoreduction in selected patient groups, reducing perioperative morbidity, saving time in preparation for the operation in patients whose performance status is not suitable for the operation or providing palliative chemotherapy, reducing mortality, and increasing the quality of life when evaluated together with emotional and cognitive functions [5]. Tissue diagnosis should usually be obtained by image-guided paracentesis, biopsy, or surgery (laparoscopy, laparotomy) before NACT can be initiated. Paracentesis, ascites cytology, and tru-cut biopsy techniques are less invasive methods for diagnosing malignancy. These techniques can be particularly useful in patients unsuitable for primary surgery and may obviate the need for open or closed surgical, diagnostic procedures.

The accuracy of ascites cytology and tru-cut biopsy was evaluated in the study. The pathology results of patients who received ascites cytology, tru-cut biopsy, ascites cytology and tru-cut biopsy simultaneously were compared with the postoperative final pathology results. The results are presented in Table 3. These results show that NACT can be administered safely to patients determined to be 'malignant' with ascites cytology and tru-cut biopsy to diagnose ovarian cancer. However, it is seen that the sensitivity for tru-cut biopsy is much higher than ascites cytology, so a tru-cut biopsy is more reliable than ascites cytology in detecting patients with ovarian cancer.

In the study of Baransi et al., including 551 patients, the sensitivity, specificity, PPV, and NPV values of ascites cytology in the diagnosis of ovarian cancer were found as 80.6%, 100%, 100%, and 16.7%, respectively [6]. Similar to the existing literature in our study; we found that the sensitivity, specificity, PPV, NPV of acid cytology were 64%, 100%, 100%, 12%, and the accuracy of the test was 65%, respectively. In the study of Zikan et al., in which 195 tru-cut biopsies including 190 patients were performed, the diagnostic accuracy was 98.3% [7]. In our study, the diagnostic accuracy of tru-cut biopsy was found to be 92%. In the study published by Vlasak et al., which included 79 patients in 2020, the only study similar to our study comparing the diagnostic reliability, accuracy, and safety of ultrasound-guided ascites cytology and tru-cut biopsy, the rates in the confirmation of malignancy were 72.9% vs. 95.8%, whereas the rates for the compliance with the final reports were 43.7% vs. 95.4%. However, not all patients included in the study had final pathology results. There are also patients whose disease progresses and cannot be operated on despite NACT [8]. False-positive cytology and biopsy results, which could lead to unnecessary NACT therapy, were not reported in any other study, including ours.

When the histopathological subtypes were evaluated in the study, the final pathology results of most patients in whom ascites cytology was reported as malignant were serous ovarian cancer. Especially in patients presenting with diffuse ascites in the abdomen, since it causes worsening in performance status due to shortness of breath and swelling, it would be appropriate to use the benefit of paracentesis to both provide relief in symptoms for the patient and to detect malignancy, especially for serous ovarian cancers.

A total of 97 ultrasound-guided minimally invasive procedures were performed in the study. The results of 92 procedures were reported because the initial materials obtained in 5 patients were not sufficient for examination. Adverse events occurred in 2 of them during the procedure. One was subcutaneous hemorrhage after tru-cut biopsy, and the other was drainage catheter-related closed-perforation in the colon. The adverse event rate was 2%. Similarly, the adverse event rate was 1% in the study of Zikan et al [7]. To achieve this rates, adverse events observed in previously published studies related to tru-cut biopsy were considered, and the procedure was performed by confirming that the patients' routine platelet and INR values were appropriate before the procedure.

In the study, the patients whose ascites cytology or tru-cut biopsy results were reported as 'malignant' were compared in terms of NACT administration, and it was observed that optimal cytoreduction rate was statistically significantly higher and hospitalization duration was shorter in the group that received NACT.

The fact that all patients had final pathology results and that we could compare simultaneous ascites cytology and tru-cut biopsy both with the final pathology and among themselves reflect the study's strength. Being a single-center study and having two pathologists evaluating all pathology reports increases the study's objectivity as well.

Study Limitations

Nevertheless, limitations include the small number of patients and its retrospective nature.

Conclusion

In conclusion, ultrasound-guided tru-cut biopsy currently appears to be the best minimally invasive method for collecting sufficient tissue samples for histopathological examination. It is also suitable for confirming recurrences, diagnosing non-gynecological malignancies infiltrating the pelvic organs, or distinguishing between malignant and benign tumors. When acid cytology can be added to this procedure, the probability of false negative results appears to be much less. However, in cases where tru-cut biopsy is not possible, ascites cytology seems to be

included in the diagnosis in appropriate patients. Through their high reliability and accuracy, these minimally invasive methods have the potential to routinely replace more invasive methods for adequate tumor sampling, such as diagnostic laparoscopy or exploratory laparotomy.

Conflict of Interest

None Declared.

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