

Ultrasonographic-guided Pervaginal Cul-de-sac Cytology in the Follow-up of Ovarian Carcinoma

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Abstract. *The aim of this study was to compare, prospectively, traditional pervaginal cul-de-sac aspiration cytology with an ultrasonographic-guided aspirate in the detection of residual or recurrent ovarian carcinoma. Patients and Methods: Fifty-one patients with ovarian carcinoma were monitored during chemotherapy (21 patients) or follow-up (30 patients) after first-line treatment. All patients underwent both traditional blind pervaginal cul-de-sac aspiration cytology and an ultrasonographic-guided pervaginal aspirate. The samples were classified as class 0 or insufficient when no mesothelial cells were detected in the aspirate. The results of cytological classification of the aspirates were compared with each other according to sampling order. Results: Samples were classified as class 0 in 56% when the traditional cul-de-sac aspiration was taken first, and in 73% when ultrasonographic-guided aspiration was taken first ($p=0.249$, Fisher's exact test). The number of class 0 samples was smaller among those taken second than among those taken first (22 (44%) vs. 33 (65%), $p=0.046$). Four recurrences were detected during the mean follow-up of six months (range 2-11 months) in 30 patients who were followed-up after the first-line treatment. In one case, a positive cul-de-sac cytology was the first and only early indication of recurrence. Conclusion: The use of ultrasonography did not improve the accuracy of the cul-de-sac aspiration. The greater amount of fluid in the cul-de-sac during the second sampling might contribute to achieving a better result.*

Ovarian cancer (OC) is the leading cause of mortality from gynecological cancers in industrialized countries. With the current treatment modalities, including surgery and platinum-taxane combination chemotherapy, 70-80% of

patients respond to therapy (1-4). However, half of the patients achieving clinical complete response are diagnosed as having persistent disease at second-look operation, and 50-60% of the patients with pathological complete response will have recurrent disease later on.

Physical examination, ultrasonography (US) (5), computed tomography (CT) (5, 6) and CA 12-5 measurement (7, 8) are the most commonly used methods for the follow-up of OC. However, the detection of primary or recurrent disease with a low tumor burden is difficult even with modern imaging techniques. Only lesions exceeding 1 cm in size can be detected by US or CT (5, 6), and diffuse peritoneal carcinosis cannot be detected even with CT (9) or positron emission tomography (PET) (10).

As OC is largely confined to the peritoneal cavity, pervaginal cul-de-sac aspiration cytology may detect otherwise occult small volume disease in cases where neither imaging techniques nor tumor marker levels correspond to the tumor load. The results of using blind cul-de-sac aspiration in the follow-up of 110 OC patients have previously been published (11). Altogether 27 recurrences were detected. Cul-de-sac aspiration cytology was the first or only indication of recurrence in one third of the cases (11). However, the high number of false-negative results, obtained in 40% to 60% of the patients with recurrence or residual disease has restricted the clinical use of this method (12, 13). The purpose of the present study was to evaluate whether the technique can be improved and the number of insufficient samples can be decreased by using ultrasonography-guided (USG) cul-de-sac aspiration.

Patients and Methods

Fifty-one patients with OC were prospectively examined during chemotherapy (21 patients) or follow-up after the treatment of primary or recurrent disease (30 patients) to evaluate their disease status. The study was performed at the Turku University Central Hospital, Finland. The study protocol was approved by the Joint Ethics Committee of the University of Turku and the University Central Hospital of Turku. All patients gave written informed consent. The follow-up included a traditional blind pervaginal cul-

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Table I. Clinical characteristics of the 51 patients with ovarian cancer.

	Mean	Median	Range
Age (years)	59	58	24-81
BMI (kg/m ²)	26.5	26.9	18.4-39.6
	No of patients		
Stage			
I		15	
II		5	
III		28	
IV		3	
Histology			
serous		29	
mucinous		6	
endometrioid		3	
clear cell		3	
anaplastic		1	
undiff. or other		9	
Grade			
1		16	
2		8	
3		26	
Undifferentiated		1	
On treatment		21	
At follow-up		30	

BMI=body mass index.

de-sac aspiration, a USG aspiration, a bimanual pelvic examination, CA 12-5 measurement and a whole abdominal CT/US (within ± one month). The clinical characteristics of the patients and the follow-up techniques are shown in Table I and Table II, respectively. The year of primary treatment ranged from 1985 to 1999. The treatment consisted of staging laparotomy and cytoreductive surgery, followed by platinum-based combination chemotherapy (96% of patients). Seven (14%) patients were also treated with radiotherapy. Two patients did not receive any postoperative therapy due to early stage disease.

The physical examination, bimanual pelvic examination, the traditional blind and USG pervaginal cul-de-sac aspirations were performed by the same investigator (MV) during 1999. USG cul-de-sac aspiration was performed with Aloka SSD 1000 (Aloka Co, Tokyo, Japan) ultrasonographic equipment and a 5-7.5 MHz transvaginal transducer with a guided puncture system was used. Alternate patients were first evaluated by traditional blind pervaginal cul-de-sac aspiration followed by USG aspiration cytology; the remaining patients were examined using the procedures in the opposite order. During cul-de-sac aspiration, the patients were placed in an anti-Trendelenburg position to accumulate peritoneal fluid in the cul-de-sac. The posterior vaginal vault was entered without any anesthesia with a 20-gauge needle attached to a syringe. Twenty ml saline was used for

Table II. Follow-up techniques used together with cul-de-sac aspiration.

	No of patients
PE	51
CA 12-5	50
Cul-de-sac aspiration	
Trad.	50
USG	51
CT/US (within ± 1 month)	26

PE= physical examination; Trad.=traditional cul-de-sac aspiration; USG=ultrasonographic-guided; CT=computed tomography; US=ultrasonography taken within ± one month from the cytologic sample.

Table III. Traditional and ultrasonographic-guided aspiration cytology results according to sampling order.

	Class 0 Number/Patients	(%)	Class I-V Number/Patients	(%)	Fisher's exact test p-value
First sample					
Trad.	14/25	(56)	11/25	(44)	0.249
USG	19/26	(73)	7/26	(27)	
Second sample					
Trad.	10/25	(40)	15/25	(60)	0.776
USG	12/25	(48)	13/25	(52)	

Trad.=traditional cul-de-sac aspiration; USG=ultrasonographic-guided cul-de-sac aspiration.

Table IV. Characteristics of the five patients with some abnormality in the follow-up examinations during chemotherapy.

No	Stage	Histology	PE	CA 12-5 U/I	Peritoneal cytology class		CT/US (within ± 1 month)
					Trad.	USG	
14	IIIC	undiff.	Pos	11	I	II	Neg
15	IIIC	serous	Pos	5	0	0	ND
18	IV	serous	Neg	753	III	I	Pos
21	IB	undiff.	Neg	5	III	III	Neg
38	IIIC	serous	Neg	132	I	I	Neg

PE=physical examination; CT=computed tomography; US=ultrasonography; Trad.=traditional; USG=ultrasonographic-guided; Pos=positive, abnormal; Neg=negative, normal; ND=not done.

irrigation. The needle-point was seen in free fluid during USG aspiration. The aspirated material was fixed in 50% ethanol, cytocentrifuged and stained with the Papanicolaou method. Cytology was evaluated according to the Papanicolaou

Table V. Characteristics of the four patients with recurrent ovarian cancer detected during follow-up.

No.	Stage	Histology	PE	CA 12-5 U/l	Peritoneal cytology class		CT/US (within ± 1 month)	Recurrent disease
					Trad.	USG		
4	IIIC	undiff.	Neg	9	0	I	Neg	8 months later at CT/US
10	IIIC	undiff.	Neg	11	I	0	ND	6 months later at CT/US
39	IIIC	serous	Neg	11	ND	0	Neg	6 weeks later, CA 12-5 elevation
50	IV	undiff.	Neg	29	IV	0	Neg	In the present study, cytology

PE=physical examination; CT=computed tomography; US= ultrasonography; Trad.=traditional; USG=ultrasonographic-guided; Neg=negative, normal; ND=not done.

classification. The samples were classified as class 0 or insufficient when there were no mesothelial cells in the aspirate. The mean follow-up time after aspiration cytology was six months (range 2-11 months).

In statistical analysis the differences between means were tested with the Student's *t*-test and cross-tabulations with Fisher's exact test.

Results

Altogether 101 cul-de-sac aspirations were taken from 51 OC patients. All patients except one were evaluated by both traditional blind pervaginal and USG cul-de-sac aspiration. In that one case, the traditional sample was not obtained due to intense pain experienced during the USG sample taken first. There were no other complications associated with the cul-de-sac aspirations. When the traditional cul-de-sac aspiration was used, 23 (46%) of the 50 patients had class I, two (4%) had class III, one (2%) class IV, and 24 (48%) class 0 or insufficient samples. In the USG aspirates, the cytological class was I in 17 (33%), II in two (4%), III in one (2%), and 0 or insufficient sample was seen in 31 (61%) of the 51 cases. There was no significant difference in the number of insufficient samples using the traditional and the USG aspiration method (*p*=0.233). The results, according to the sampling order, are presented in Table III. Among the samples taken second, the number of insufficient or class 0 samples was smaller than among the samples taken first (22 vs. 33, *p*=0.046). Patient characteristics, consisting of BMI, the number of chemotherapy courses and intra-abdominal adhesions detected at primary operation, did not correlate with the number of class 0 samples.

Five out of the 21 patients monitored during chemotherapy had abnormal findings detected with some of the follow-up techniques. These results are shown in Table IV.

Four recurrences were detected during the mean follow-up time of six months (range 2-11 months) in the 30 patients who were followed-up after the primary or second-line treatment (Table V). In one case, class IV peritoneal cytology was the first and only early indication of recurrence.

Discussion

Peritoneal fluid can easily be obtained by cul-de-sac aspiration and the method is familiar to most gynecologists. Analysis of peritoneal aspiration cytology is the only commonly available outpatient method for the detection of occult disease in cases where small volume tumor is not detected by imagine techniques, or when tumor marker values do not correspond to the tumor load. A diagnostic finding in a peritoneal aspirate could even decrease the need for more expensive and invasive methods of follow-up (14). However, the high number of false-negative results, in 40% to 60% of the patients with recurrence or residual disease (11-13), has restricted the clinical use of this method. In the present study, we evaluated whether the number of insufficient samples can be reduced with USG cul-de-sac aspiration.

When we compared the samples taken first, *i.e.*, samples that are comparable in clinical practice, using either the traditional blind method or USG aspiration, the USG pervaginal aspiration did not reduce the number of insufficient samples. The number of class 0 samples was even slightly higher when USG sampling was used, although the difference was not statistically significant. When all blindly taken and USG samples were compared, the number of insufficient samples was also slightly higher if the sample was taken using US guidance. There is no obvious reason for this unexpected finding. The amount of free peritoneal fluid in the cul-de-sac is very small in clinically disease-free patients. Since the bowel loops are directly above the vaginal vault, seeing them during the USG aspiration may increase the fear of perforating the bowel with the aspiration needle. Hence the procedure might be performed overcautiously, a fact which may increase the number of insufficient samples.

There were fewer insufficient samples among the aspirations taken second, regardless of which method had been used. Thus, the greater amount of fluid in the cul-de-

sac during the second sampling may contribute to achieving a better result.

One patient experienced the USG aspiration as painful, but none of the patients had bleeding, infection, or any other complications. Therefore, both techniques can be considered safe. The traditional blind pervaginal cul-de-sac aspiration proved to be at least as reliable as the USG aspiration and is simpler to perform. Therefore, it can be recommended as the primary technique for obtaining a peritoneal sample for cytological evaluation during the follow-up of OC.

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