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The Length of Peritoneal Surgical Manipulation Correlates with Serum CA 125 Levels

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Abstract. *Background: Peritoneal recovery after uncomplicated serum manipulation usually lasts 7 days and high values of serical CA 125 are measured following abdominal surgery. The aim of this study was to assess a possible correlation between peritoneal manipulation and serical CA 125 levels following abdominal surgery for benign diseases. Patients and Methods: Twenty-eight patients with abdominal benign disease were operated on. They were pooled into three groups of low, intermediate and high peritoneal manipulation, according to the extent of laparotomy and length of surgical peritoneal manipulation. Venous blood samples (5 ml) were taken from each patient 24-48 hours before surgery, 12-24 hours after surgery and on the 4th and 7th postoperative day. CA 125 levels were quantified by microparticle enzyme immunoassay. Results: After surgery, patients having high peritoneal manipulation showed significantly higher levels of CA 125 compared to the preoperative levels. In particular, the length of peritoneal manipulation was correlated with increasing levels of the marker (p<0.0001). Conclusion: Peritoneal manipulation was significantly correlated to serum CA 125 levels; therefore its role as marker of peritoneal surgical injury should be considered.*

Cancer antigen 125 (CA 125), a high molecular-weight glycoprotein of 200,000 Da, is the antigenic determinant recognised by the murine monoclonal antibody OC-125 as described by Bast *et al*. (1) CA 125 neoplastic marker has been demonstrated to be useful in detecting and predicting primary neoplasms or recurrences of the female genital tract, and in particular this antigen is expressed in nearly 80% of patients with epithelial ovarian cancer (2-7). However, elevated CA 125 serum levels are also observed in other clinical conditions such as pleural and pericardial effusions, dialysis, cirrhosis of the liver with ascitic fluid, peritoneal tuberculosis, and in patients who undergo abdominal surgery for benign disease (8-15).

Peritoneal cells have been demonstrated to be a valid source of CA 125 (16-17) and are very highly produced in areas of inflammation (18). In the context that peritoneal repair after uncomplicated surgery has been demonstrated to be complete approximately on the 7th postoperative day (19), this study investigated whether the length of peritoneal manipulation is related to postoperative CA 125 blood levels in patients who have undergone abdominal surgery during the immediate postoperative period.

Patients and Methods

Eligibility criteria. Patients who had undergone surgery for abdominal diseases were enrolled in this prospective observational study. Other inclusion criteria were: no evidence of neoplastic disease, pleural effusion or ascitic fluid, normal preoperative liver and renal function. All patients gave informed consent for their participation in the study, which was approved by the local Ethics Committee.

Patients and study design. Twenty-eight consecutive patients underwent surgery for benign abdominal disease. Patient characteristics are reported in Table I. Patients were analysed and stratified according to the extent of laparotomy (in centimetres) and length of surgical peritoneal manipulation (in minutes) as follows: patients submitted to <8 cm laparotomy extension and <30 minutes of peritoneal manipulation (group 1, low manipulation), patients submitted to <8 cm laparotomy extension and 30-90 minutes of peritoneal manipulation (group 2, intermediate mani-pulation) and patients with >8 cm laparotomy extension and >90 minutes of peritoneal manipulation (group 3, high manipulation). Manipulation of the peritoneum was calculated starting from the time of incision of peritoneal surface until finishing at the closure of abdominal fascia, which completes the procedure. Patients were operated on under general or spinal anaesthesia. Surgical procedures performed

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Table I. *Patients and surgical procedures*.

Pt no.	Age (years)	Gender	Diagnosis	Surgical procedure	Peritoneal manipulation period (minutes)
1	58	M	Inguinal hernia	Hernioplasty with prosthetic mesh	15
2	91	M	Inguinal hernia	Hernioplasty with prosthetic mesh	15
3	77	M	Inguinal hernia	Hernioplasty with prosthetic mesh	10
4	77	M	Inguinal hernia	Hernioplasty with prosthetic mesh	5
5	72	M	Inguinal hernia	Hernioplasty with prosthetic mesh	10
6	53	M	Inguinal hernia	Hernioplasty with prosthetic mesh	5
7	75	M	Inguinal hernia	Hernioplasty with prosthetic mesh	5
8	63	M	Bilateral inguinal hernia	Bilateral hernioplasty with mesh	10
9	72	M	Inguinal hernia	Hernioplasty with prosthetic mesh	5
10	71	M	Inguinal hernia	Hernioplasty with prosthetic mesh	5
11	66	M	Inguinal hernia	Hernioplasty with prosthetic mesh	5
12	54	F	Cholelithiasis	Laparoscopic cholecystectomy	60
13	80	M	Cholelithiasis	Laparoscopic cholecystectomy	60
14	67	F	Umbelical hernia and intestinal necrosis	Hernioplasty and resection	90
15	70	M	Cholelithiasis	Laparoscopic cholecystectomy	55
16	78	F	Gangrenous cholecystitis	Laparoscopic cholecystectomy	85
17	45	F	Cholelithiasis	Laparoscopic cholecystectomy	55
18	87	F	Perforated duodenal ulcer	Laparotomy and suture	30
19	74	M	Cholelithiasis	Laparoscopic cholecystectomy	70
20	41	M	Cholelithiasis	Laparoscopic cholecystectomy	65
21	68	F	Cholelithiasis	Laparoscopic cholecystectomy	35
22	42	F	Cholelithiasis	Laparoscopic cholecystectomy	65
23	73	M	Laparotomical hernia	Hernioplasty with prosthetic mesh	90
24	83	M	Bilateral iliaco-femoral aneurysm	Aorto-bifemoral bypass	390
25	66	M	Aorto-bisiliac aneurysm	Aorto-bisiliac bypass	240
26	73	M	Abdominal aortic aneurysm	Aorto-bisiliac bypass	150
27	61	M	Cholelithiasis	Laparotomic cholecystectomy	100
28	57	M	Cholelithiasis and choledocholithiasis	Laparoscopic cholecystectomy and choledocotomy	125

Table II. *CA 125 serum levels at different times (mean±SD)*.

CA 125 level	Group 1 (11 pts)	Group 2 (12 pts)	Group 3 (5 pts)
Preop. 24-48 hours	12.39±8.78*	11.36±3.92**	8.89±2.63***
Postop. 12-24 hours	11.70±7.50	9.51±3.59	10.24±4.09
Postop. 4th day	-	15.36±8.11	35.30±17.60
Postop. 7th day	11.82±7.06*	33.75±20.49**	73.57±35.99***

*p= not significant; **p<0.001; ***p<0.008

are reported in Table I. One or two abdominal drainages were placed in 10/12 patients of the second group, in 5/5 patients of the third group and removed at 24 to 96 and 72 to 96 hours postoperatively, respectively. All surgical specimens were submitted for histological examination.

Biochemical assessment. Venous blood samples (5 ml) were taken from each patient 24-48 hours before surgery, 12-24 hours after surgery and on the 4th and 7th postoperative day. CA 125 concentrations in the sera were determined by microparticle enzyme immunoassay (MEIA), using OC-125 monoclonal antibody, AxSYM CA 125 assay (ABBOTT Laboratories, Diagnostic Division, Abbott Park, IL, USA). The normal value of CA 125 was considered to be less or equal than 35 UI/ml in this study.

Statistical analysis. Data are given as the mean±standard deviation (SD). Preoperative and postoperative CA 125 levels within each group and among the different groups of patients were compared by using one or two-tailed Student's *t*-test. Linear regression analysis was used to correlate postoperative CA 125 levels with length of peritoneal manipulation.

Results

Surgery. No patient showed evidence of cancer during surgery or on histological analysis. Mean values of operating time, extension of laparotomy and length of peritoneal manipulation were measured. Patients were pooled as follows: 11 patients submitted to <8 cm laparotomy extension and <30 minutes of peritoneal manipulation (group 1, low manipulation), 12 patients submitted to <8 cm laparotomy extension and 30-90 minutes of peritoneal manipulation (group 2, intermediate manipulation) and 5

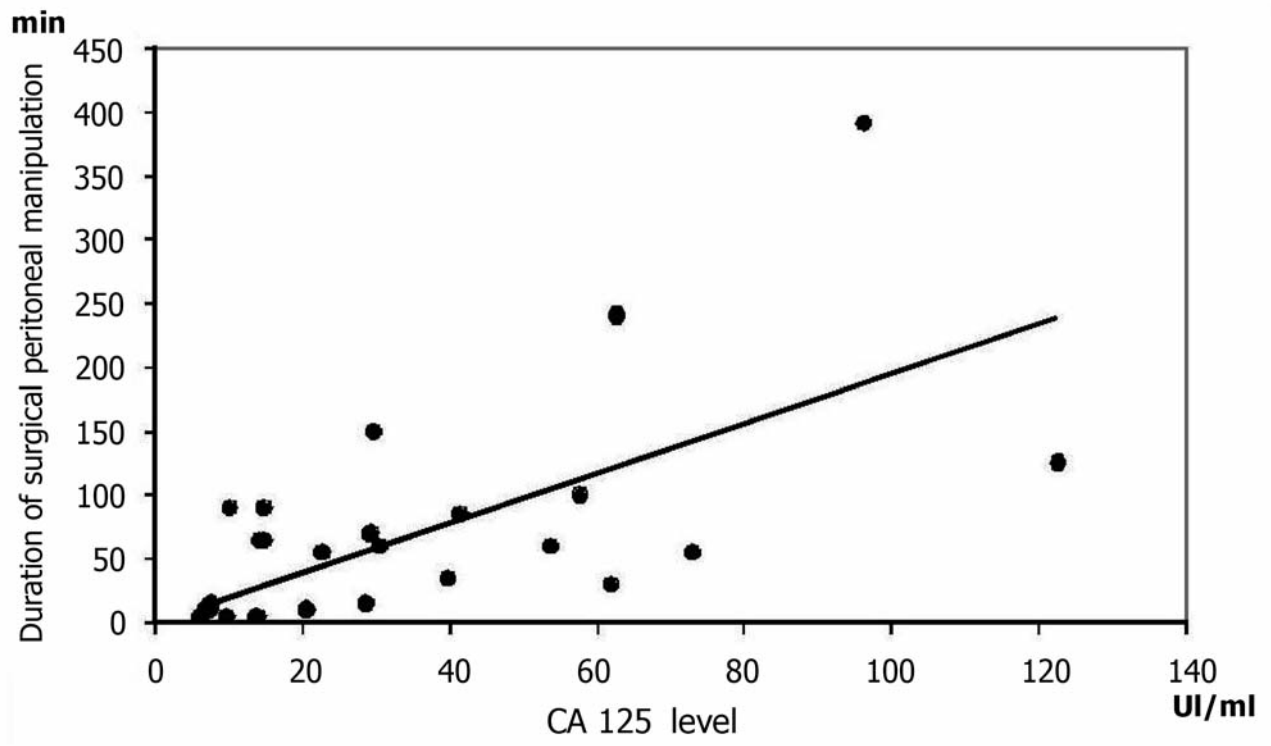

Figure 1. *Correlation between CA 125 serum levels and duration of surgical peritoneal manipulation. r=0.6394, p<0.0001.*

patients with >8 cm laparotomy extension and >90 minutes of peritoneal manipulation (group 3, high manipulation).

No intraoperative or postoperative deaths occurred. Five postoperative complications occurred in 5 patients: 1 wound seroma and 2 wound Hematomas (first group) and 2 pneumonia (second group). All complications were treated conservatively and no further peritoneal manipulations were performed.

CA 125 levels. The mean±SD levels of serical CA 125 of the three groups are reported in Table II. In the first group of patients, at different times, no significant modification of CA 125 levels occurred. In the second group (intermediate manipulation), decreasing CA 125 levels were observed immediately after the operation ($p<0.001$), followed by increasing values on the fourth and seventh postoperative day ($p<0.05$ and $p<0.001$ respectively). In the third group (high manipulation), progressive increasing CA 125 levels were observed starting from the preoperative to the seventh postoperative day ($p<0.008$). In 11/12 patients (second group) and in 5/5 (third group), the highest CA 125 level was measured on the 7th postoperative day.

There was a statistically significant correlation between the CA 125 values on the 7th postoperative day and the corresponding length of peritoneal manipulation, a ($r=0.6394$, $p<0.0001$; Figure 1).

Discussion

CA 125 is a useful but not specific tumour marker that is used worldwide for the clinical evaluation of patients with epithelial gynaecological disease (19-21) and for predicting peritoneal dissemination in patients with gastric cancer (23) or advanced non-Hodgkin's lymphoma (24-25). The low specificity of CA 125 arises from the fact that it is produced in presence of ascitic fluid, by benign ovarian cysts, by cancer (26) and peritoneal cells (17-18).

Peritoneal repair after surgery has been extensively studied and it has been demonstrated that cases of uncomplicated surgery the peritoneal repair process, is usually completed 7 days after surgery (19). Peritoneal repair correlates with postoperative CA 125 increasing levels, (24) therefore suggesting its role as a marker of mesothelial membrane irritative processes (13). Peritoneal manipulation strongly correlates with CA 125 secretion; its serical levels resulted significantly higher in 19 patients submitted to abdominal surgery compared with 21 patients operated for extra-abdominal disease, even if only one blood sample was considered, 48 hours after surgery, and also some patients with cancer were enrolled (17).

In order to describe the serical CA 125 secretion profile occurring following peritoneal manipulation in uncomplicated surgery, its levels in patients were monitored during 7

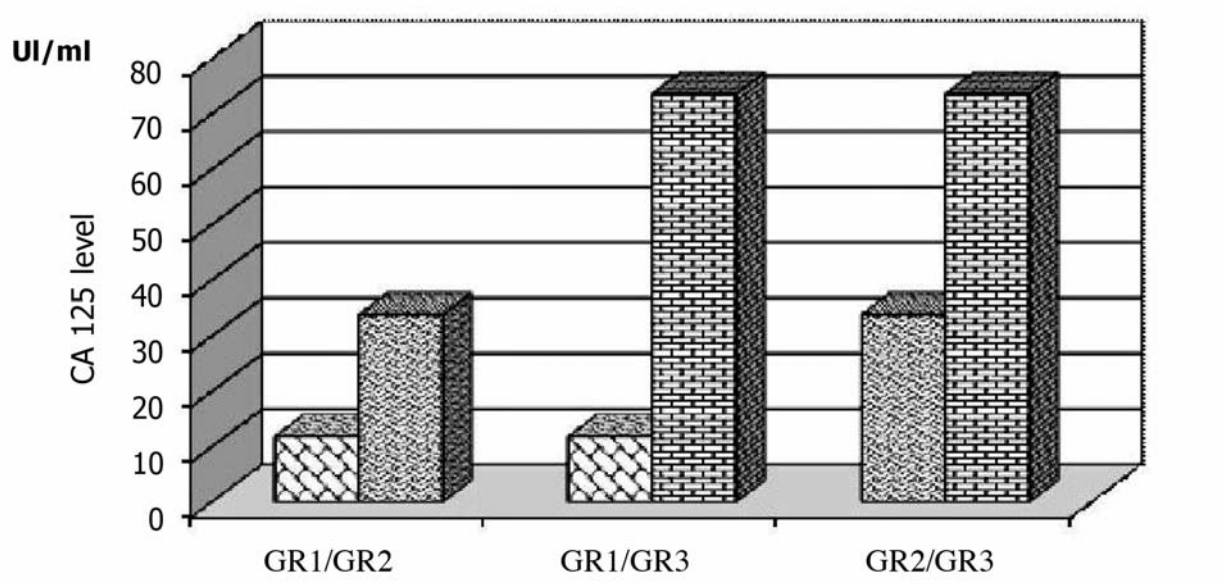

Figure 2. *Evaluation of serical CA 125 in the three groups of patients on the seventh postoperative day. GR 1=GROUP 1; GR 2=GROUP 2; GR 3=GROUP 3; GR1 vs. GR2: p<0.001, GR1 vs. GR3: p<0.00001, GR2 vs. GR3: p<0.006.*

postoperative days. Three peritoneal manipulation times were considered: short (group 1, less than 30 minutes), intermediate (group 2, between 30 and 90 minutes) and prolonged (group 3, more than 90 minutes), according to the mean length of the peritoneal manipulation usually required during the surgical procedures performed.

The results of the current study demonstrate that preoperative CA 125 levels were not statistically different among the 3 groups of patients. Furthermore, according to another report (12), on the 7th postoperative day, the maximal CA 125 levels were significantly different among the three groups (see Figure 2) and the length of peritoneal manipulation correlated with the increasing marker levels (see Figure 1).

Zeimet *et al*. stated that human mesothelial cells are one of the most potent sources of CA 125 in the serum and its release is five times higher than from the ovarian cancer cells (18). Considering that CA 125 has half-life of 5 days, and that the highest levels were measured after 7 days, it can be supposed that it was continuously secreted during the whole 7-day postoperative period that was investigated.

Peritoneal cell secretion of CA 125 is activated in presence of cytokines released from the system of lymphocytes, macrophages, fibroblastic and endothelial cells when peritoneal trauma or flogistic processes occur (27-28). Furthermore, CA 125 production has been found to be significantly enhanced when peritoneal cells were cultured in the presence of proinflammatory cytokines such us recombinant interleukin 1 (rIL-1), tumour necrosis factor-alpha (TNF-alpha), interferon gamma (IFN-gamma) and lipopoly- saccharide from *Escherichia coli* (16,18). During abdominal surgery, peritoneal injury following trauma or flogistic processes has been demonstrated to be the major cause of adhesion formation and, in their turn, adhesions are frequently involved in bowel obstruction, chronic abdominal pain and infertility (29, 32). Since serum CA 125 level is correlated to peritoneal injury, its early measurement during the post- operative period might help to identify those patients who are at high risk of adhesion formation.

In conclusion, there is an early significant *in vivo* peritoneal secretion of CA125 in patients who undergo abdominal surgical procedures for benign disease. The length of peritoneal reaction following surgical peritoneal injury may be revealed by the CA 125 serum levels and this marker may be useful for the prediction of the risk of developing continuous inflammatory peritoneal reaction which is, with angiogenesis, cell migration, proliferation and tissue remodelling, one of the major contributing factors in abdominal adhesion growth after surgical operations.

References

1 Bast RC, Feeney M, Lazarus H, Nadler LM, Colvin RB and Knapp RC: Reactivity of monoclonal antibody with human ovarian carcinoma. J Clin Invest *68*: 1331-1337, 1981.

2 Jacobs I and Bast RC: The CA-125 tumor associated antigen: a review of the literature. Hum Reprod *4*: 1-12, 1989.

3 Van der Zee AG, Duk J, Aalders JG, BoontjeAH, Ten Hoor KA and De Bruijn HW: The effect of abdominal surgery on the serum concentration of the tumour-associated antigen CA-125. Br J Obstet Gynecol *97*: 934-938, 1990.

4 Kenemans P, Yedema CA, Bon GG and Von Mensdorff-Pouilly S: CA 125 in gynecological pathology: a review. Eur J Obstet Gynecol *49*: 115-124, 1992.
5 Yedema CA, Kenemans P and Thomas CM: CA 125 serum levels in the early post-operative period do not reflect tumour reduction obtained by cytoreductive surgery. Eur J Cancer *29*: 966-971, 1993.
6 Fisken J, Leonard RC, Stewart M, Beattie GJ and Sturgeon C: The prognostic value of early CA 125 serum assay in epithelial ovarian carcinoma. Br J Cancer *68*: 140-145, 1993.
7 Peters-Engl C, Obermair A, Heinzl H, Buxbaum P, Sevelda P and Medl M: CA 125 regression after two completed cycles of chemotherapy: lack of prediction for long-term survival in patients with advanced ovarian cancer. Br J Cancer *81*: 662-666, 1999.
8 Talbot RW, Jacobsen DJ, Nagorney DM, Malkasian GD and Ritts RE: Temporary elevation of CA 125 after abdominal surgical treatment for bening disease and cancer. Surg Gynecol Obstet *168*: 407-411, 1989.
9 Molina R, Filella X, Bruix J, Mengual P, Bosch J and Calvet X: Cancer antigen 125 in serum and ascitic fluid of patients with liver diseases. Clin Chem *37*: 1379-1383, 1991.
10 Ho-dac-Pannekeet MM, Hiralall JK, Struijk DG and Krediet RT: Markers of peritoneal mesothelial cells during treatment with peritoneal dialysis. Adv Perit Dial *13*: 17-22, 1997.
11 Jimenez C, Diaz C, Selgas R, Auxiliadora Bajo M, Del Peso G and Sanchez-Tomero JA: Peritoneal kinetics of cancer antigen 125 in peritoneal dialysis patients: the relationship with peritoneal outcome. Adv Perit Dial *15*: 36-39, 1999.
12 Harkki-Siren P, Sjoberg J, Toivonen J and Tiitinen A: Clinical outcome and tissue trauma after laparoscopic and abdominal hysterectomy: a randomized controlled study. Acta Obstet Gynecol Scand *79*: 866-871, 2000.
13 Sevinc A, Buyukberber S, Sari R, Kiroglu Y, Turk HM and Ates M: Elevated serum CA-125 levels in hemodialysis patients with peritoneal, pleural or pericardial fluids. Gynecol Oncol *77*: 254-257, 2000.
14 Mas MR, Comert B, Saglamkaya U, Yamanel L, Kuzhan O and Kuzhan O: Ca-125; a new marker for diagnosis and follow-up of patients with tuberculous peritonitis. Dig Liver Dis *32*: 595-597, 2000.
15 Kawanishi H, Moriishi M, Harada Y, Sakikubo E, Nagai T and Tsuchiya S: Necessity of correcting cancer antigen 125 appearance rates by body surface area. Adv Perit Dial *16*: 22-25, 2000.
16 Zeillemaker AM, Verbrugh HA, Hoynck van Papendrecht AA and Leguit P: CA 125 secretion by peritoneal mesothelial cells. J Clin Pathol *47*: 263-265, 1994.
17 Epiney M, Bertossa C, Weil A, Campana A and Bischof P: CA 125 production by the peritoneum: *in-vitro* and *in-vivo* studies. Hum Reprod *15*: 1261-1265, 2000.
18 Zeimet AG, Marth C, Offner FA, Obrist P, Uhl-Steidl M and Feichtinger H: Human peritoneal mesothelial cells are more potent than ovarian cancer cells in producing tumor marker CA-125. Gynecol Oncol *62*: 384-389, 1996.
19 diZerega GS and Campeau JD: Peritoneal repair and post-surgical adhesion formation. Human Reproduction Update *6*: 547-555, 2001.
20 Meier W, Stieber P, Hasholzner U, Gropp M and Fateh Moghadam A: Prognostic significance of CA125 in patients with ovarian cancer and secondary debulking surgery. Anticancer Res *17(4B)*: 2945-2947, 1997.
21 Walach N and Gur Y: Leukocyte alkaline phosphatase, CA15-3, CA125 and CEA in cancer patients. Tumori *84*: 360-363, 1998.
22 Sarandakou A, Phocas I, Botsis D, Sikiotis K, Rizos D and Kalambokis D: Tumour-associated antigens CEA, CA125, SCC and TPS in gynaecological cancer. Eur J Gynaecol Oncol *19*: 73-77, 1998.
23 Nakata B, Chung KH, Kato Y, Yamashita Y, Maeda K and Onoda N: Serum CA 125 level as a predictor of peritoneal dissemination in patients with gastric carcinoma. Cancer *83*: 2488-2492, 1998.
24 Pabst T and Ludwing C: CA 125: a tumor marker in non-Hodgkin lymphomas? J Clin Oncol *13*: 1827-1828, 1995.
25 Ozguroglu M, Turna H, Demir G, Doventas A, Demirelli F and Mandel NM: Usefulness of the epithelial tumor marker CA 125 in non-Hodgkin's lymphoma. Am J Clin Oncol *22(6)*: 615-618, 1999.
26 Bergmann JF, Bidart JM, George M, Beaugrand M, Levy VG and Bohuon C: Elevation of CA 125 in patients with benign and malignant ascites. Cancer *59*: 213-217, 1987.
27 Ohzato H, Yoshizaki K and Nishimoto N: Interleukin-6 as a new indicator of inflammatory status: detection of serum level of interleukin-6 and C-reactive protein after surgery. Surgery *111*: 201-209, 1992.
28 Ellstrom M, Bengtsson A, Tylman M, Haeger M, Olsson JH and Hahlin M: Evaluation tissue trauma after laparoscopic and abdominal hysterectomy: measurements of neutrophil activation and release of interleukin-6, cortisol and C-reactive protein. J Am Coll Surg *182*: 423-430, 1996.
29 Mrstik M, Kotseos K, Ma C and Chegini N: Increased expression of interferon-inducible protein-10 during surgically induced peritoneal injury. Wound Rep Reg *11(2)*:120-126, 2003.
30 Chegini N: Peritoneal molecular enviroment, adhesion formation and clinical implication. Front Biosci *7*: 91-115, 2002.
31 Ozel H, Avsar FM, Topaloglu S and Sahin M: Induction and assessment methods used in experimental adhesion studies. Wound Rep Reg *13(4)*: 358-364, 2005.
32 Holmdahl L and Ivarsson LM: The role of cytokines, coagulation and fibrinolysis in peritoneal tissue repair. Eur J Surg *165*: 1012-1019, 1999.

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