

Selected oxidative stress markers in gynecological laparoscopy

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Videosurgery Miniinv 2015; 10 (1): 92–100

DOI: 10.5114/wiitm.2014.47449

Abstract

Introduction: The surgical stress response after laparoscopy is smaller when compared with open surgery, and it is expected that after minimally invasive surgery the possible development of oxidative stress will be less severe.

Aim: To evaluate markers of pro-oxidant activity – levels of lipid peroxides and malondialdehyde – and activity of the antioxidant enzymes superoxide dismutase and glutathione peroxidase in the perioperative period in patients undergoing gynecological laparoscopy and to determine whether the duration of laparoscopy can affect these changes.

Material and methods: The study included 64 patients, divided into two groups: group 1 with duration of laparoscopy up to 20 min, and group 2 with duration of the operation over 40 min. Blood samples were collected before anesthesia, 5 min after release of pneumoperitoneum, and 10 h after surgery.

Results: A statistically significant increase in the levels of lipid peroxides and malondialdehyde in samples collected after surgery was found in comparison with values obtained before surgery. Also statistically significant differences existed between groups of patients with different duration of surgery. Superoxide dismutase and glutathione peroxidase activity values were significantly decreased. They were also significantly different between the two groups with different duration of surgery.

Conclusions: In our study, levels of the markers of pro-oxidant activity increased and levels of the markers of antioxidant enzymes decreased, suggesting development of oxidative stress. The duration of laparoscopic procedures affects the severity of the presented changes.

Key words: gynecological laparoscopy, duration of laparoscopy, oxidative stress.

Introduction

Each operation is an injury that causes a hormonal, metabolic, immune and hemodynamic imbalance [1]. They create an image of the surgical stress response [2]. Injury, septic shock and surgical stress lead to increased production of free radicals [3]. And surgical stress can be associated with increased levels of lipid peroxides. It was confirmed by Szymczyk *et al.* [4] with women after abdominal hysterectomy.

This may be associated with activation of polymorphonuclear neutrophils generating superoxide anion. Abe *et al.* [5] observed release of active forms of oxygen in the early postoperative period by neutrophils. Deitch *et al.* [6] postulated that the source of reactive oxygen species (ROS) in operative stress may be the activation of xanthine oxidase, persisting in his studies until the first day after surgery. Excessive production of free radicals and decreased

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activity of antioxidant mechanisms lead to damage of macromolecules and disorders of cell metabolism, the state described as oxidative stress.

The factors responsible for the rise of oxidative stress during laparotomy are the manipulation of intestines, decreased perfusion in the visceral area and the formation of an ischemia-reperfusion syndrome, injury of the peritoneum, and inflammatory cell stimulation [7]. Less trauma to the peritoneum, and intestines or formation of pneumoperitoneum may cause differences in the generation of postoperative oxidative stress.

In recent years there have been reports of oxidative stress in the perioperative period in patients undergoing laparoscopy. Most of these concern surgical and urological operations and experimental studies carried out on animals.

In the available literature, there are very few reports on oxidative stress during gynecological laparoscopy. Usually cited is the work of Zhang *et al.* [8], as well as Aran *et al.* [9].

Aim

Assessment of pro-oxidative mechanism markers (malondialdehyde (MDA) and lipid peroxides) and activity of antioxidant systems (superoxide dismutase (SOD) and glutathione peroxidase) during gynecological laparoscopy. Determination of the relationship between the duration of the operation and the level of the studied parameters.

Material and methods

The study was conducted in a group of 83 patients who underwent surgery in the Department of Gynecology and Obstetrics of F. Raszeja Hospital in Poznan. It obtained ethical approval from the Bioethics Committee of the Medical University of Poznan (Resolution No. 262). The study involved women with operational risk ASA I/II. The study excluded patients burdened with additional diseases,

and taking medication or hormonal contraceptives within 2 months prior to the surgery. Persons under 20 and over 30 years of age, and with body mass index (BMI) over 25 kg/m², were excluded.

In the chosen group indications for surgery included cases of primary infertility (19), secondary infertility (14), pelvic pain syndrome (10), a suspected pelvic endometriosis (19), and ovarian cysts (21). During the operation, in 4 cases it was necessary to proceed to laparotomy. After the operation, patients were divided into groups according to the length of laparoscopy (up to 20 min, from 21 to 40 min, more than 40 min), and 11 patients with the duration of the operation between 21 and 40 min were excluded from further investigations. In 1 case fever occurred on the first day after the surgery. Also in 3 cases, not all laboratory tests were performed. Finally, the results of tested samples obtained from 64 patients divided into 2 groups were analyzed (Table I).

All patients were operated on by the same operating team. Patients were premedicated in the evening and 1 h before the surgery with midazolam 0.1 mg/kg. The anesthesia was started with propofol 1.5–2.5 mg/kg, vecuronium 0.08–0.1 mg/kg, fentanyl 2.5–4.0 mg/kg. General anesthesia was used in a semi-closed system, with monitoring of electrocardiogram, non-invasive blood pressure, saturated pressure of arterial oxygen, heart rate, end tidal carbon dioxide, capnography curve, the concentration of anesthetic gases, and neuromuscular transmission (NMT). Anesthesia was continued with 1 MAC of sevoflurane, fentanyl 2.5–4.0 mg/kg, and vecuronium 0.05 mg/kg. For ventilation, a mixture of air with oxygen (FiO₂ = 0.4) was used. Laparoscopy was performed using laparoscopic sets of Aesculap and RZ-Medizintechnik. Aesculap insufflators established intraperitoneal pressure of 13 mm Hg. When replacing tools or aspirating, gas flows were increased so that pressure fluctuations did not exceed ±1 mm Hg. Three or four trocars were pierced in conventional places. The electro-

Table I. Characteristics of patients

Group	Group 1 (< 20 min)	Group 2 (> 40 min)
Number of patients, <i>n</i>	33	31
Age of patients, median (range) [years]	25 (21–28)	25 (20–28)
BMI, median (range) [kg/m ²]	19.7 (18.3–21.2)	20.2 (18.7–22.0)
Duration of operation, median (range) [min]	19 (15–20)	43 (41–50)

surgical set consisted of mono- and bipolar instruments, with an argon module and vessel sealing system. Tissues were removed from the abdominal cavity with extraction bags. Duration of laparoscopy was counted from the moment of puncture with a Veress needle until removing the last trocar and elimination of pneumoperitoneum.

During the laparoscopy lasting less than 20 min (group 1), chromoscopy was performed to assess the patency of the fallopian tubes. Cauterization of the ovaries was also done, single endometriosis foci were coagulated, and mild adhesions were freed.

During the laparoscopy lasting over 40 min (group 2), numerous adhesions were freed and endometriosis foci were coagulated. In case of fallopian hydrosalpinx, salpingostomy was performed. Paraovarian cysts were excised. Theca lutein cysts and ovary cystic teratomas were enucleated.

Sampling

Blood was collected into heparinized 6 ml tubes three times: 1) immediately prior to anesthesia, 2) immediately after the operation (5 min after the release of pneumoperitoneum), 3) 10 h after the operation, while performing routine blood tests.

The collected samples were centrifuged at 3500 rpm at 4°C. The resulting plasma was frozen, and 0.9% NaCl was added to the cells in the ratio 1 : 1; these were then mixed and centrifuged three times at 3500 rpm for 10 min at 4°C. Washed red cells were divided into two samples, and distilled water in a 1 : 1 ratio was added. In plasma the levels of lipid peroxides and MDA were determined. In the hemolysate, activity of glutathione peroxidase and SOD was determined.

Determination of malondialdehyde concentration in plasma

To determine the concentration of MDA in plasma, the BIOXYTECH LPO-586 (Oxis International Inc., USA) test was used. The product of the test is a stable chromophore whose absorbance is measured spectrophotometrically at a wavelength of 586 nm.

Calculating the absorbance of test samples was preceded by measuring the absorbance of standard samples with known and increasing concentrations of MDA. A standard curve was plotted according to the manufacturer. For final calculation of the concentration of MDA in the sample, the pattern recom-

mended by the producer was used. The results were expressed in micromoles per liter.

Determination of the concentration of lipid peroxides in plasma

To determine the lipid peroxide level in the plasma, the BIOXYTECH LPO-560 (Oxis International Inc., USA) test was used. This test uses a colorimetric method based on the ability of peroxide to oxidize ferrous ions (Fe^{2+}) to ferric ions (Fe^{3+}), and, combined with the color indicator xylenol orange, form a complex whose absorbance is measured with a spectrophotometer at a wavelength of 560 nm. Adding the substance reducing lipid peroxides to part of the sample allowed the creation of a blank. The concentration of lipid peroxides was calculated from the formula recommended by the manufacturer. The results were expressed in micromoles per liter.

Determination of superoxide dismutase (EC.1.15.1.1.) in red blood cells

To determine the activity of SOD in red blood cells, the spectrophotometric assay BIOXYTECH SOD-525 (Oxis International Inc., USA) was used. In this test the property of SOD increasing the autoxidation rate of 5,6,6a,11b-tetrahydro-3,9,10-trihydrobenzofluorene in basic medium to form a chromophore was used. The absorbance of the chromophore was measured at a wavelength of 525 nm. The rate of reaction for the blind and test samples was calculated on the basis of a standard curve, taking into account the absorbance change per unit of time. Determination of SOD activity using the ratio V_s/V_c is possible due to the supplied test table or on the basis of the calculations according to the manufacturer's formula. The results were expressed as U/g Hgb.

Determination of glutathione peroxidase (EC 1.11.1.9) in red blood cells

Glutathione peroxidase (GSH-Px) activity in red blood cells was measured by the Ransel test (Randox Laboratories Ltd., UK). This test uses the method of determination of GSH-Px developed by Paglia and Valentine. The change in the absorbance of each sample was determined in 1-minute intervals for 4 min at a wavelength of 340 nm at 37°C using a spectrophotometer. For the calculations, the high-

est increase in absorbance subtracted with the result obtained for the blank was used. The enzyme activity was presented in U/g Hgb.

Statistical analysis

All results were checked for compliance with the Gaussian distribution by the Shapiro-Wilk test, and compliance with the normal distribution was confirmed. For each experimental group, comparisons between groups were performed “before”, “immediately after”, and “after 10 h”, using the ANOVA test for repeated measures with comparisons between groups (pairwise comparison). Comparisons between different experimental groups in selected time periods were made using Student’s *t* test with the level of significance at $p < 0.05$.

Results

Level of malondialdehyde

The level of MDA (Table II) in group 1 before surgery was $3.396 \pm 0.146 \mu\text{mol/l}$, and increased significantly to $4.383 \pm 0.143 \mu\text{mol/l}$ immediately after and to $4.203 \pm 0.137 \mu\text{mol/l}$ at 10 h after the surgery. The level of MDA before laparoscopy in group 2 was $3.406 \pm 0.136 \mu\text{mol/l}$ and increased significantly after the operation and 10 h after laparoscopy to $6.366 \pm 0.147 \mu\text{mol/l}$, and $5.970 \pm 0.121 \mu\text{mol/l}$, respectively. A statistically significant difference be-

tween groups 1 and 2 in both samples taken after the surgery was also demonstrated.

Level of lipid peroxides

Levels of lipid peroxides showed a similar direction of change (Table III). In group 1 the value of $8.106 \pm 0.122 \mu\text{mol/l}$ increased to $10.037 \pm 0.167 \mu\text{mol/l}$ immediately after the operation and to $9.304 \pm 0.131 \mu\text{mol/l}$ at 10 h after surgery. Also, there was a significant difference between the last two results. The level of lipid peroxides in group 2 before laparoscopy was $8.103 \pm 0.136 \mu\text{mol/l}$ and significantly increased immediately after, and at 10 h after surgery to $12.796 \pm 0.174 \mu\text{mol/l}$, and to $10.810 \pm 0.211 \mu\text{mol/l}$, respectively. The latter two results and post-operative values between groups 1 and 2 also differed significantly.

Activity of superoxide dismutase

The activity of SOD in red blood cells of patients in group 1 before the surgery was $2543.1 \pm 91.7 \text{ U/g Hb}$ (Table IV). The activity decreased to $2027.1 \pm 67.9 \text{ U/g Hb}$ immediately after surgery and remained at $2122.3 \pm 91.3 \text{ U/g Hb}$ at 10 h after surgery; these changes were significant. The SOD activity in red blood cells of patients in group 2 before the operation was $2516.2 \pm 66.7 \text{ U/g Hb}$, and decreased to $1567.4 \pm 101.7 \text{ U/g Hb}$ immediately after surgery and to $1917.2 \pm 82.1 \text{ U/g Hb}$ at 10 h after surgery,

Table II. Level of MDA in the plasma of patients undergoing gynecological laparoscopy

Group	Group 1 (< 20 min) [$\mu\text{mol/l}$]	Group 2 (> 40 min) [$\mu\text{mol/l}$]
Before operation	3.396 ± 0.146	3.406 ± 0.136
Immediately after operation	$4.383^* \pm 0.143$	$6.366^{***} \pm 0.147$
Ten hours after operation	$4.203^{**} \pm 0.137$	$5.970^{***} \pm 0.121$

Results are presented as mean \pm SD. Group 1 – $n = 33$, group 2 – $n = 31$. *Statistically significant differences compared with the values before surgery at $p < 0.001$. **Statistically significant differences compared with the values after the operation at $p < 0.001$. ***Statistically significant differences compared to the values in group 1 at $p < 0.001$. Selected oxidative stress markers in the gynecological laparoscopy.

Table III. Level of lipid peroxides in the plasma of patients undergoing gynecological laparoscopy

Group	Group 1 (< 20 min) [$\mu\text{mol/l}$]	Group 2 (> 40 min) [$\mu\text{mol/l}$]
Before operation	8.106 ± 0.122	8.103 ± 0.136
Immediately after operation	$10.037^* \pm 0.167$	$12.796^{***} \pm 0.174$
Ten hours after operation	$9.304^{**} \pm 0.131$	$10.810^{***} \pm 0.211$

Results are presented as mean \pm SD. Group 1 – $n = 33$, group 2 – $n = 31$. *Statistically significant differences compared with the values before surgery at $p < 0.001$. **Statistically significant differences compared with the values after the operation at $p < 0.001$. ***Statistically significant differences compared to the values in group 1 at $p < 0.001$.

Table IV. Activity of SOD in red blood cells of patients undergoing gynecological laparoscopy

Group	Group 1 (< 20 min) [U/g Hb]	Group 2 (> 40 min) [U/g Hb]
Before operation	2543.1 ±91.7	2516.2 ±66.7
Immediately after operation	2027.1* ±67.9	1567.4*** ±101.7
Ten hours after operation	2122.3* ±91.3	1917.2*** ±82.1

Results are presented as mean ± SD. Group 1 – n = 33, group 2 – n = 31. *Statistically significant differences compared with the values before surgery at $p < 0.001$. **Statistically significant differences compared with the values after the operation at $p < 0.001$. ***Statistically significant differences compared to the values in group 1 at $p < 0.001$.

Table V. Activity of glutathione peroxidase in red blood cells of patients undergoing gynecological laparoscopy

Group	Group 1 (< 20 min) [U/g Hb]	Group 2 (> 40 min) [U/g Hb]
Before operation	35.07 ±1.22	34.11 ±1.83
Immediately after operation	28.67* ±1.88	22.17*** ±2.26
Ten hours after operation	29.20* ±1.39	25.89*** ±1.87

Results are presented as mean ± SD. Group 1 – n = 33, group 2 – n = 31. *Statistically significant differences compared with the values before surgery at $p < 0.001$. **Statistically significant differences compared with the values after the operation at $p < 0.001$. ***Statistically significant differences compared to the values in group 1 at $p < 0.001$.

with statistical significance. The results obtained after the surgery in groups 1 and 2 also differed significantly.

Activity of glutathione peroxidase

The GSH-Px activity in red blood cells of patients in group 1 before the surgery was 35.07 ±1.22 U/g Hb (Table V), decreased statistically significantly to 28.67 ±1.88 U/g Hb immediately after, and remained at the level of 29.20 ±1.39 U/g Hb at 10 h after the operation. In patients from group 2 activity of GSH-Px before the operation was 34.11 ±1.83 U/g Hb. Peroxidase activity significantly decreased to a value of 22.17 ±2.26 U/g Hb immediately after surgery and remained at a level of 25.89 ±1.87 U/g Hb 10 h after the surgery. Postoperative values of group 1 and 2 also differed significantly.

Discussion

Professor Kurt Semm from Kiel started in the 1980s the era of laparoscopic gynecological operations throughout Europe and the United States of America. Also in 1983, as a gynecologist, he made the first laparoscopic appendectomy [10]. For patients it is the preferred method compared to the open surgical approach or natural orifice transluminal endoscopic surgery (NOTES) [11]. The advantage

of laparoscopy is less stress than in the open operative techniques [8]. One of the major components of the operative stress is oxidative stress [12]. Laparoscopic technique, being less traumatic, is characterized, according to most authors, by less severe oxidative stress than the open technique used in the same procedure [12–14].

Our research, based on the model of gynecological laparoscopy, concerned groups differing in duration of surgery. In our study we found an increase in the level of lipid peroxides and MDA, in the samples taken 5 min after the release of pneumoperitoneum and 10 h after the surgery. The activity of antioxidant enzymes SOD and GSH-Px were decreased. This direction of change was observed in both groups of patients. Our results can be compared with a few reports on oxidative stress in patients undergoing surgery for gynecological reasons. These include the work of Taskin *et al.* [15], who carried out laparoscopic operations of ovarian tumors. They found decreasing activity of antioxidant enzymes (SOD, catalase and GSH-Px) and glutathione levels in homogenates of the peritoneum slices. Zhang *et al.* [8], in turn, in patients with uterus fibroids, found increased pro-oxidative activity 5 min after the release of pneumoperitoneum. The activity of GSH-Px and total antioxidant activity in this study showed a slight decrease. Changes in the activity

of antioxidant systems were also seen 24 h after the surgery. The probable cause of these changes, according to the author, was the ischemia-reperfusion syndrome. These results correlate with those obtained in our study and indicate the development of oxidative stress in the course of laparoscopy, performed at high pressure pneumoperitoneum.

Aran *et al.* [9] in patients undergoing gynecological laparoscopy observed an increase in the level of ischemia-modified albumin, treating it as an exponent of oxidative stress. In that study the level of MDA did not increase, which was possibly related to the treatment of the sample taken at 10 min after anesthesia as a control.

Further discussion on pro-oxidant activity can be found in the works concerning laparoscopic surgery by Bukan *et al.* [12], Glantzounis *et al.* [16], and Zengin *et al.* [17]. All the authors cited above obtained, in samples taken after the release of pneumoperitoneum, results in line with ours. Evaluation of pro-oxidant activity at 12 h, a period similar to that in our work, was conducted only by Zengin *et al.* [17], and the results are compatible with ours.

Reduction of SOD and GSH-Px, as in our study, in the course of laparoscopic cholecystectomy was demonstrated by Olakowski *et al.* [18]. Stipančić *et al.* [19], in turn, using the same model in the blood plasma analyzed total antioxidant status (TAS) level and SOD activity on the first, third and seventh post-operative day. They found no changes in the level of TAS, and an increase in SOD activity on the 7th day after surgery, compared to the preoperative period.

Some reports dealing with the development of oxidative stress in the course of laparoscopy are based on experimental studies. Antioxidant behavior of the system in the course of experimental laparoscopy was analyzed by Akbulut *et al.* [20], who noted a decrease in SOD activity after a laparoscopic donor nephrectomy. Similarly Cevrioglu *et al.* [21] confirmed the decrease in the activity of glutathione reductase and the level of reduced glutathione in erythrocytes and peritoneum of rats. The cause of reduced activity of antioxidant enzymes in the course of laparoscopy, which is an example of ischemia-reperfusion syndrome, can be, according to Sun *et al.* [22], the reduction of their synthesis, consumption in the processes of removing free radicals or deactivation by excess of reactive oxygen species. In the course of laparoscopy, one of the factors that can stimulate pro-oxidative mechanisms is the gas

fed to produce pneumoperitoneum. Carbon dioxide is most often used for this purpose. However, according to Wong *et al.* [23] it causes acidosis, which can stimulate the local inflammatory response in the peritoneum [24]. This supposition is confirmed by the study of Redmond *et al.* [25]. Pross *et al.* [26] observed increased lipid peroxidation in lung tissue. It was related to increased migration and activation of neutrophils, which reached a maximum at 18 h after the end of the pneumoperitoneum. Neutrophil activation may be one of the factors responsible for the persistent increased levels of MDA and peroxides in our study at 10 h after the surgery. In order to eliminate the adverse effects of CO₂ in the course of laparoscopy, other gases were tested, especially helium [27, 28]. Shuto *et al.* [28] obtained similar results using both CO₂ and helium, and stated that hemodynamic changes depend on the value of the intraperitoneal pressure, and not on the type of gas used.

Glantzounis *et al.* [29] observed the development of oxidative stress in patients undergoing laparoscopic cholecystectomy, and drew attention to the role of ischemia-reperfusion syndrome in the generation of free radicals. Experimental studies on animals showed that peritoneal edema increases the intraperitoneal pressure, resulting in ischemia of abdominal organs. This, in turn, increases the formation of free radicals [30]. The occurrence of abdominal ischemia was confirmed in humans during laparoscopic cholecystectomy [31]. A thorough discussion of abdominal organs ischemia under pneumoperitoneum can also be found in the work of Schäfer and Krähenbühl [32]. All cited authors agree as to the occurrence of ischemia in abdominal organs during pneumoperitoneum. Guven *et al.* [33] in experimental studies observed a reduction in flow within the ovary with subsequent development of oxidative stress.

Ischemia caused by pneumoperitoneum is transient. After the release of pneumoperitoneum normal tissue perfusion is restored [34]. Therefore, laparoscopy should be considered as an example of ischemia-reperfusion syndrome.

Zweier *et al.* and Ambrosio *et al.* [35, 36] noted an increase of free radical generation in 10–20 s of reperfusion. Other authors have demonstrated peak generation of free radicals in the 5th [37] and in the 15th min of reperfusion [38]. In the early period of reperfusion, even in the reduced oxygen availability,

in vascular endothelial cells increased O_2 conversion to a reactive form is observed, with a severity proportional to the depth of hypoxia [39].

Glantzounis *et al.* [40], summarizing the reports on changes in the liver in the course of ischemia-reperfusion syndrome, stated that activation of neutrophils releases ROS [41]. However, the main source of ROS in ischemic tissues during reperfusion is a xanthine oxidase (XO) system [42]. There is a consensus that during laparoscopy ischemia-reperfusion syndrome is growing at an average level, and there is an increase in XO activity in the gut [43]. There are cases of intestinal necrosis during prolonged laparoscopic procedures, which Hasson *et al.* [44] associate with critical ischemia, rarely observed in patients with good general condition.

In our study, one of the objectives was to determine whether the duration of laparoscopy may correlate with the severity of oxidative stress. The obtained results indicate that the short-term procedure is combined with a benign course of oxidative stress. Peroxide and MDA levels were statistically higher in patients whose operation time exceeded 40 min, compared with the short-term laparoscopy. Dismutase activity in samples taken immediately after the operation was lower in patients operated on for over 40 min. A statistically significant decrease in GSH-Px activity was also noted in this group of patients. In the available literature there are no studies using a similar model and analyzing these markers.

The available literature lacks reports indicating the scope of laparoscopy performed and the type of operating procedures, mainly indicating the duration of the ischemia-reperfusion syndrome and more broadly the effect of duration of pneumoperitoneum on the activity of pro- and antioxidant mechanisms.

Quite useful for the interpretation of the development of oxidative stress in our study may be a correlation between the duration of laparoscopy and the severity of ischemia-reperfusion syndrome, the main source of free radicals. Unsal *et al.* [45], during experimental laparoscopy, found a dependence of severity of any ischemia-reperfusion syndrome on both the intraperitoneal pressure and the duration of pneumoperitoneum. Similarly, Schilling *et al.* [46], at a constant intraperitoneal pressure, observed progressive reduction of the flow in the visceral area. Skorzyński *et al.* [47] published the results of hemodynamic parameters such as the speed of blood flow in the descending aorta, total peripheral vascular

resistance, and stroke volume in patients undergoing gynecological laparoscopy. The procedures were divided into three groups: lasting up to 30 min, from 30 to 60 min, and over 60 min. Statistically significant differences were found in the results obtained during the medium- and long-term laparoscopy compared to short-term laparoscopy. This confirms the importance of pneumoperitoneum duration for the formation of hemodynamic disorders. Emir *et al.* [48] noted an increase of xanthine oxidase activity in the large intestine of animals undergoing laparoscopy, 20 min after pneumoperitoneum was established. Zweier *et al.* [35] believe that there is a correlation between the severity of oxidative stress and the duration of ischemia, which can lead to inactivation of antioxidant enzymes. This was also corroborated by Porreca *et al.* [49], who reported an increase in the activity of catalase and GSH-Px in an ischemia-reperfusion model of myocardial infarction, in which rats were subjected to 10 min of ischemia followed by 30 min of reperfusion. Extending the ischemic time in this model led to reduced activity of the enzymes.

In the available literature, two opinions concerning the relationship between the deepening of oxidative stress and the duration of laparoscopy are presented. Polat *et al.* [50] stated that the amount of free radicals produced during pneumoperitoneum depends not only on the severity of ischemia-reperfusion injury, but also on its duration. A different point of view is presented by Urena *et al.* [51], who analyzed the level of isoprostanes in the urine of patients undergoing urological laparoscopy. Urological operations are performed, however, without pneumoperitoneum, and the formation of isoprostanes depends on sources other than the formation of MDA and peroxides. Our results indicate that shorter duration of the laparoscopy is associated with less severe oxidative stress. However, further studies are necessary to analyze the role of pneumoperitoneum duration and the types of laparoscopic procedures for the development of oxidative stress.

Conclusions

In the course of gynecological laparoscopy the development of oxidative stress is manifested by increased levels of lipid peroxides and MDA and reduced activity of the antioxidant enzymes SOD and glutathione peroxidase. Degree of oxidative stress correlates with the duration and extent of laparoscopic surgery.

Conflict of interest

The authors declare no conflict of interest.

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Received: 2.10.2014, **accepted:** 26.10.2014.