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Original Research

Subclinical activation of coagulation and fibrinolysis in laparoscopic cholecystectomy: Do risk factors exist?

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ABSTRACT

Purposes: This study examines whether inherent patient-related risk factors (age, gender) modify the effect of laparoscopic cholecystectomy (LC) upon the coagulation and fibrinolysis cascades.
Methods: This observational study included 119 low-risk for deep vein thrombosis (DVT) patients undergoing elective LC, without thromboprophylaxis. Pre-operatively and 24 h post-operatively we measured PT–INR, aPTT, FDP, d-dimer, and fibrinogen. Color Doppler scan of the lower extremity was performed the 1st post-operative day. Differences before and after surgery were analyzed with respect to risk factors.
Results: No clinically or ultrasound evident DVT was observed. INR (1.04 ± 0.06 vs. 1.12 ± 0.11, $p < 0.0001$), d-dimer (0.38 ± 0.36 vs. 0.9 ± 0.64, $p < 0.0001$), plasma fibrinogen (380.8 ± 74.9 vs. 403.8 ± 78.8, $p = 0.0001$) and FDP positivity exhibited statistically significant increase after surgery. The levels of aPTT did not exhibit any significant change. Concerning d-dimer, older age was associated with higher pre-operative concentrations; older patients accordingly exhibited more intense increase in d-dimer and FDP positivity after surgery. Male sex was associated with higher PT–INR and aPTT before surgery, as well as with more pronounced increase in PT- INR postoperatively; similarly, older age was associated only with higher PT–INR before surgery.
Conclusions: Despite no DVT, significant increase in PT–INR, d-dimer, FDP and fibrinogen appeared after LC. This may be attributed to surgical trauma and pneumoperitoneum effects on the portal vein flow. Elderly subjects and males seem particularly vulnerable, demonstrating more sizeable changes.

1. Introduction

Three major risk factors have been described for Deep Vein Thrombosis (DVT) during Laparoscopic Cholecystectomy (LC): surgical trauma, pneumoperitoneum causing inferior vena cava compression and the anti-Trendelenburg position resulting in venous stasis in the lower limbs. The definitive clinical diagnosis of DVT is difficult, traditionally requiring the use of ascending venography, an invasive technique, now replaced by Colour Doppler Ultrasound.[1] Clinical scoring systems such as Well's clinical pretest score[2] and newer ones incorporating d-dimer assay have been created in order to stratify the risk for DVT in medical patients so as to limit the use of expensive imaging studies.[3–5] Lack of systematic studies focusing in surgical patients limits the use of such algorithms in this population.

Pharmaceutical and mechanical DVT prophylaxis has been supported by some authors and current NICE clinical guidelines for all patients undergoing LC.[6,7] Nevertheless according to other authors the role of DVT prophylaxis is less defined in laparoscopic surgery,[8–10] as DVT incidence after LC is extremely rare.[10,11]

A number of studies have investigated changes in the coagulation and fibrinolytic cascades accompanying LC. D-dimer exhibits a well established increase post-operatively[12–22] even in the presence of Low Molecular Weight Heparin (LMWH) prophylaxis.[16,17,19] Similarly a significant increase in plasma fibrinogen levels is demonstrated post-operatively.[13,15,17–21] Concerning Prothrombin Time (PT) and its International Normalized Ratio (INR),[12,15,19–21] activated Partial Thromboplastin Time (aPTT)[12,15,18–20] controversial results have appeared in the literature.

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This study examines the coagulation and fibrinolytic response after LC in a group of low-risk patients for DVT, assessing PT–INR, aPTT, d-dimer and Fibrin/Fibrinogen Degradation Products (FDP) and fibrinogen. This study makes a step beyond the existing knowledge, examining whether inherent patient-related risk factors (age, gender) modify the effect of LC upon the coagulation and fibrinolysis cascades.

2. Methods

2.1. Study sample

This observational study included 119 low-risk for DVT (<1 on the clinical model proposed by Wells[2,3]) patients undergoing elective LC from February 2005 to January 2006. All patients were white caucasian with age range 29–86 years. Exclusion criteria were: history of coagulation disorder, malignancy, hepatic failure, past DVT or pulmonary embolism, superficial venous insufficiency, conversion of the operation to open cholecystectomy, anticoagulation therapy (heparin, Low Molecular Weight Heparin, Coumadin) and antiplatelet medication (COX inhibitors, ADP receptor inhibitors) if not stopped at least five days prior to hospital admission.

Informed consent was obtained by all study participants. This study has been approved by the local Institutional Review Board.

2.2. Surgical procedure

Each patient underwent uncomplicated LC under general anesthesia. A standard operative and anesthesia protocol was followed by all surgeons; the experience of the operating surgeon, as demonstrated by the past number of LCs performed, was recorded. With the patient in the Trendelenburg position a Veress needle through a small incision under the umbilicus was used to insufflate with CO_2 until pneumoperitoneum of 12 mmHg was achieved. The patient was then placed to anti-Trendelenburg position with a left tilt throughout the operation. Standard American school positioning of the surgical team and four ports were used. Intraoperative cholangiogram was not performed. No patient received prophylactic LMWH nor had compressive stockings during or after the surgery. Patients were allowed free fluids by mouth, and ambulation was encouraged in the evening of surgery. Patient discharge took place, routinely, 24–48 h after the operation.

2.3. Patient evaluation

Pre-operatively the patients underwent complete medical examination and past medical history was obtained; sex and age were recorded. Blood samples were drawn to measure PT, also expressed as INR, aPTT, FDP, d-dimer, and plasma fibrinogen. The above were measured with an automated STA® analyzer from Diagnostica Stago (France). INR, given its standardization as well as its inclusion in well established hepatic scoring systems (MELD, Child-Pugh) was used in parallel with PT. Qualitative and semi-quantitative determination of FDP in Plasma by Latex Agglutination was performed with the use of FDP PLASMA kit. The STA® - Neoplastine® CI plus kit was used for PT and INR determination (normal range 12–15 s and 0.9–1.2) and the STA® – PTT A 5 kit for aPTT determination (normal range 28–40 s). The STA® - fibrinogen 5 kit was used for fibrinogen evaluation (normal range 200–400 mg/dL). Immuno-Turbidimetric assay of d-dimer with the STA® - Liatest® D-DI kit was performed (normal <0.5 mg/L).

Post-operative evaluation included clinical re-evaluation using the Wells Clinical Prediction Rule for DVT.[2,3] A new blood sample was obtained the morning of the first post-operative day for PT, aPTT, FDP, d-dimer, and fibrinogen assessment. All patients underwent a Color Doppler Ultrasound scan of the lower extremities on the 1st post-operative day to rule out subclinical DVT. Patient follow-up took place at our outpatient department on the 8th post-operative day for clinical signs of DVT.

2.4. Statistical methods

PT, INR, aPTT, d-dimer and fibrinogen were treated as continuous variables, whereas FDP positivity was treated as a binary variable (0 = negative, 1 = positive). To assess whether the above parameters exhibited statistically significant changes before and after surgery, Wilcoxon matched-pairs signed-ranks test and Chi-square test for paired observations were appropriately performed.

Subsequently, in case the change was proven statistically significant, the respective change was calculated for each of the above parameters and denoted as Δ(parameter). For instance, Δ(PT) = (PT after surgery) − (PT before surgery). Regarding the binary FDP positivity variable, a comparable but distinct approach was adopted, as "*FDP positivity emerging after surgery*" denoted patients who developed FDP positivity solely after surgery, being negative before surgery; the opposite (i.e. patients being FDP positive before surgery and converting to FDP negative after surgery) was not observed in the studied sample (see below).

To examine whether possible risk factors (sex, age) were associated with pre-surgery concentration, as well as with Δ(parameter) and "FDP positivity emerging after surgery", non-parametric statistics (Mann-Whitney-Wilcoxon test for independent samples and Spearman's rank correlation coefficient), Chi-square and Fisher's exact test were appropriately performed. The statistical test performed is indicated in the text. Statistical analysis was performed with STATA 8.0 statistical software (Stata Corp, College Station, TX, USA).

3. Results

The features of the study sample are summarized in Table 1. No clinically evident DVT occurred in any of our patients post-operatively. Color Doppler Ultrasound scan showed no signs of DVT; no patient was found having clinical signs of DVT at the 8th day follow-up.

Table 2 presents the changes in the measured parameters before and after surgery. PT, INR, d-dimer, plasma fibrinogen and FDP positivity exhibited statistically significant increase after surgery; on the contrary, the levels of aPTT did not exhibit any statistically significant change. More specifically, with respect to FDP, two patients exhibited positivity before surgery; those patients remained positive after surgery and eleven additional patients exhibited FDP positivity post-operatively.

Table 3 presents the associations between pre-operative measurements, as well as changes in measured parameters, with possible risk factors. Male sex was associated with higher PT and INR before surgery, as well as with more pronounced increase in PT and INR; similarly, older age was associated only with higher PT and INR before surgery. Concerning d-dimer, older age was associated with higher pre-operative concentrations; older patients accordingly exhibited more intense increase in d-dimer values after surgery. Older age appeared as a predictor of emerging FDP positivity after surgery, but not of higher pre-operative FDP positivity. Regarding aPTT, the positive correlation between its baseline values and male sex is worth reporting; nevertheless, given that the changes in aPTT were not statistically significant, no putative associations with risk factors were sought.

4. Discussion

A distinct feature of this study is the inclusion of DVT low-risk patients. Given the rarity of DVT in LC,[10,11] DVT incidents occurred neither at clinical nor at ultrasonographic level. It should be stressed that elective LC as a minimal access surgical procedure bears low-risk for DVT.[8,10] In spite of the favorable prognostic factors in this sub-population a number of authors[6,7,16,17,19] propose or investigate the use and effects of pharmaceutical DVT prophylaxis; as a result significant variability in current practice has been demonstrated.[8,9] In this context our study agrees with the evidence that DVT low-risk patients may undergo LC without DVT prophylaxis as suggested by previously published literature,[8,10] but essentially with early ambulation.

Interestingly enough in this low-risk subgroup a significant activation of both coagulation and fibrinolysis cascades became evident. This observation is in accordance with existing data, and has been attributed to factors such as surgical trauma and related inflammatory response, pneumoperitoneum, and the anti-Trendelenburg

Table 1
Description of the study sample.

Continuous variables	Mean ± SD
Age (years)	57.6 ± 14.6
Experience of the surgeon (number of procedures)	1328 ± 695
Categorical variable	Percentage (N)
Sex	
Male	41.2% (49/119)
Female	58.8% (70/119)

Table 2
Measured parameters before and 24 h after surgery (mean ± SD).

Measured parameters	Before surgery	24 h after surgery	p
d-dimer (mg/L)	0.38 ± 0.36	0.90 ± 0.64	<0.0001[a]
Fibrinogen (mg/dL)	380.8 ± 74.9	403.8 ± 78.8	0.0001[a]
FDP positivity	1.7% (2/119)	10.9% (13/119)	0.0009[b]
95%CI	0.2–5.9%	5.9–18.0%	
PT (sec)	13.5 ± 0.6	14.3 ± 1.0	<0.0001[a]
INR	1.04 ± 0.06	1.12 ± 0.11	<0.0001[a]
aPTT (sec)	32.6 ± 3.0	33.2 ± 4.3	0.2757[a]

[a] p-value derived from Wilcoxon matched-pairs signed-ranks test.
[b] p-value derived from chi-square test for paired observations.

position.[12–22]. The significant post-operative thrombosis/fibrinolysis activation (including increase in d-dimer levels) in the absence of clinically significant DVT suggests that d-dimer may not have optimal screening and predictive properties in LC patients.

D-dimer and fibrinogen were statistically higher 24 h post-operatively. Interestingly their mean post-operative values were above the upper limit of the normal range (i.e. d-dimer <0.5 mg/L and fibrinogen 200–400 mg/dL). Post-operative d-dimer elevation has been reported constantly in several studies, with[16,17,19] or without[12–15,18,20–22] administration of DVT pharmaceutical prophylaxis. Accordingly fibrinogen elevation has been documented, irrespectively of the administration of DVT prophylaxis.[13,15,17–21] D-dimer, measured after LC, is shown to rise after surgery, peak at 24 h and remain elevated for at least 1 week after surgery.[22,23] It has been proposed that surgical trauma causes the activation of thrombosis and fibrinolysis. It seems that a correlation exists between the intensity of the surgical trauma and the post-operative values of d-dimer and fibrinogen. Indeed, a series of studies comparing open cholecystectomy with LC demonstrated a more pronounced increase in the former.[13,17,19,20]

Our results demonstrated statistically significant increase in PT–INR 24 h after surgery. This is in accordance with studies reporting significant[19] or slight increase[12,15,24]; on the other hand, we are in discrepancy with reports documenting no difference[20] or even decrease in PT – INR.[21] Our finding may be inscribed into a wider context, that of alterations in hepatic function during LC,[24,25] attributed to reduced portal vein flow due to pneumoperitoneum.[24] It would be tempting to speculate that the above mechanism of PT – INR prolongation might be favorable in reducing the risk of DVT in LC.

Regarding inherent risk factors, patient age emerged as an important parameter. Older patients presented with higher d-dimer levels, PT and INR; indeed, positive associations between age and d-dimer have been reported in healthy individuals,[26] whereas decrease in prothrombin activity has also been described in the elderly.[27] More importantly, however, older age was associated with a more pronounced post-operative increase in d-dimer and FDP levels; in other words older patients, apart from beginning with higher starting d-dimer levels manifest an additional, more intense, activation of the fibrinolysis cascade. To our knowledge this is the first time that such a finding has been reported, and may warrant special consideration on this particularly vulnerable sub-population.

In our analysis gender appeared to mediate interesting effects. Male patients demonstrated higher baseline PT – INR and aPTT values; higher aPTT[28] and decreased prothrombin activity[27] has been observed earlier in male subjects. Noticeably males exhibited a consistently distinct profile concerning INR; a more sizeable increase in INR was noted post-operatively in addition to the already higher baseline. Nevertheless the clinical significance of this observation remains obscure, given its limited magnitude and the lack of a clear underlying biochemical explanation.

Our study bears certain limitations. The Wells clinical model for the prediction of DVT has been developed upon and used in medical outpatients. However it is a well established scoring system and at the time of the study design, the Caprini risk stratification system[27] was not widely used. Regarding the set of risk factors, the inclusion of additional surgeon- and operation- related parameters such as total operation, anesthesia and pneumoperitoneum time, would be desirable; nevertheless the adoption of a standard protocol may point to the fact that no significant bias was introduced. In addition inclusion of further inherent risk factors such as smoking habits, BMI, physical activity etc might yield more elaborate information about vulnerable subgroups. It should also be noted that if one takes into account the current thromboprophylaxis guidelines as issued by NICE[6] and the Caprini scoring system[29] some of our patients would be considered to be at risk for DVT because of their age (>60 years old). At any case at this point this study is the first to document that inherent risk factors, as age and gender, are capable of modifying the coagulation and fibrinolytic response during LC.

In conclusion, in our low-risk sample no DVT sequelae were observed despite the non administration of pharmaceutical

Table 3
Associations with possible risk factors (mean ± SD).

Parameters before surgery	d-dimers	p^a	Fibrinogen	p^a	FDP positivity emerging after surgery	p^b	PT	p^a	INR	p^a	aPTT	p^a
Sex		0.905		0.861		1.0		0.034		0.002		0.0005
Male	0.36 ± 0.23		380.3 ± 72.1		2.0% (1/49)		13.7 ± 0.7		1.06 ± 0.08		33.7 ± 3.1	
Female	0.40 ± 0.43		381.2 ± 77.3		1.4% (1/70)		13.4 ± 0.4		1.02 ± 0.04		31.8 ± 2.7	
Age (years)		0.024		0.781		0.412		0.014		0.003		0.974
≤70	0.36 ± 0.41		377.6 ± 76.0		1.3% (1/76)		13.5 ± 0.5		1.03 ± 0.06		32.4 ± 2.7	
>70	0.43 ± 0.24		388.5 ± 77.5		4.4% (1/23)		13.7 ± 0.6		1.06 ± 0.07		32.5 ± 4.1	
Changes in measured parameters	**Δ(d-dimers)**	**p^a**	**Δ(Fibrinogen)**	**p^a**	**FDP positivity emerging after surgery**	**p^c**	**Δ(PT)**	**p^a**	**Δ(INR)**	**p^a**	**N/A**	**N/A**
Sex		0.471		0.279		0.762		0.009		0.022		
Male	+0.57 ± 0.58		+30.2 ± 79.0		10.2% (5/49)		+0.97 ± 0.78		+0.10 ± 0.10			
Female	+0.47 ± 0.48		+17.9 ± 60.5		8.6% (6/70)		+0.63 ± 0.77		+0.07 ± 0.08			
Age (years)		0.007		0.262		0.009		0.781		0.987		
≤70	+0.41 ± 0.43		+17.4 ± 65.3		6.6% (5/76)		+0.73 ± 0.78		+0.08 ± 0.09			
>70	+0.84 ± 0.75		+26.4 ± 88.4		26.1% (6/23)		+0.85 ± 0.92		+0.09 ± 0.10			

[a] p-value derived from Mann-Whitney-Wilcoxon test for independent samples.
[b] p-value derived from Fisher's exact test.
[c] p-value derived from chi-square test; N/A not applicable, since aPTT did not exhibit any statistically significant changes.

prophylaxis. Significant increase in PT– INR, d-dimer, FDP and fibrinogen levels were noted at 24 h after LC. Elderly subjects seem particularly vulnerable, as they demonstrated marked d-dimer upregulation and PT–INR prolongation.

Conflict of interest

None declared.

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The research did not receive funding from any source outside our hospital.

Ethical approval

Observational prospective study. No Ethical Approval required. Approval by the scientific board of the hospital.

Author contribution

Dimitrios Ntourakis is the paper author and study designer.

Theodoris N Sergantanis is the paper co-auther and performed statistical analysis.

Ioannis Georgiopoulos did important data collection.

Eleni Papadopoulou did data collection and performed the radiology investigation.

Lambros Liasis did data collection.

Emmanuel Kritikos did data analysis and contributed to the study design.

Periklis Tzardis did data analysis and corrected the paper.

Vasilios Laopodis corrected and approved the paper.

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