Peer Review File

I read the manuscript with interest, as I have myself looked into this disease. There are, however, some issues that merit attention; my comments follow, according to each section of the text.

ABSTRACT
Line 13: Raw numbers are required here, not only percentages.

Response: Thank you for this comment. Accordingly, we have inserted the raw numbers in the Abstract.

Changes in the text: See page 1, lines 15–17.

Line 18: The knowledge that testicular salvage mainly depends upon symptom duration is more than consolidated. In my opinion, including this analysis will distract the reader from the main point of the paper.

Response: Thank you for this comment. We agree with your opinion and have thus deleted the relevant sentence in the Abstract.

Changes in the text: See page 1, line 18.

INTRODUCTION
Line 3: The problem is the word "sudden". In general urologic practice, the time during which pain develops is a clinical criterion in the differential diagnosis of acute testicular pain. Removing the word solves the problem.

Response: Thank you for this comment. Accordingly, we have deleted this word in the Introduction.

Changes in the text: See page 3, line 3.

Line 4: The 1:4000 incidence is universally replicated -- in my opinion it is outdated, we have better data now.

Response: Thank you for this comment. Accordingly, we have amended our description in the Introduction.

Changes in the text: See page 3, lines 4–5.

Line 12: Care is needed here. What we usually see is that it is not patient cooperation, or its lack thereof, the source of US Doppler difficulties in the diagnosis of the disease -- my observations (not only mine...) point to technical limitations of the individual performing/interpreting the exam.

Response: Thank you for this valuable comment. We agree with your point that there may be differences in how ultrasonography is performed and how the results are interpreted among doctors. Accordingly, we have amended the Introduction.

Changes in the text: See page 3, lines 13–16.

MATERIAL AND METHODS
Line 11-12: We require knowing how many patients were excluded and the reasons -- studies like this are already plagued by selection bias, so there is no need to further augment the problem. A flowchart with the study population breakdown would be quite interesting here.

Response: Thank you for this comment. We agree that a flowchart could be helpful in understanding the breakdown of the study population. We have amended the Methods and have provided a flow diagram of the study design (Figure 1).

Changes in the text: See page 5, lines 9–10; page 21, line 1; and Figure 1.

Statistical analysis
Retrospective studies with opportunistic datasets like this one rely heavily on previous assumptions and model choice. It does not suffice to say "multivariate analysis" -- this is simply too vague. Also, why means and SDs. Much of the data will not be normally distributed...

Response: Thank you for this comment. Variables with p values < 0.25 in the univariate analysis were included in the multivariate model. We used the "enter" method (Purposeful selection of variables in logistic regression. Bursac Z, et al. Source Code Biol Med. 2008. PMID: 19087314). We also agree with your second comment. However, if the sample size is more than 25, we can assume that the continuous variables will follow a normal distribution (Homogeneity of variance in the two-sample means test; Moser BK & Stevens GR; The American Statistician. 1992;46:19; Estimating the mean and variance from the median, range, and the size of a sample. Hozo SP, et al. BMC Med Res Methodol. 2005. PMID: 15840177). Therefore, we conducted the statistical analysis as described in the Methods.

RESULTS
Line 6: No difference in age between patients with inflammation versus torsion? Really? This absence of difference suggests (even more to us) selection bias.

Response: Thank you for this comment. Studies have reported that inflammation can occur at any age, whereas testicular torsion occurs at a relatively young age. In this study, we evaluated only patients younger than 25 years. This could have affected the results regarding the non-difference in age between the testicular torsion and epididymitis groups. According to your comment, we have amended the limitations section of the Discussion.

Line 16 (next page): Seasonal variation in the disease's incidence (torsion) has been shown in many places -- in all continents, as a matter-of-fact. It is wrong for the authors to say that there were no statistical differences in this matter, as it seems that this statistical analysis was inadequate. These lines

could be omitted, I think.

Response: Thank you for this comment. Accordingly, we have deleted the descriptions and Supplementary Figure 1, which previously showed the seasonal variation of the disease in the manuscript.

Changes in the text: See page 9, line 4 and page 13, line 2.

DISCUSSION
Line 12: 10% survival after 6 hours? A broader review of the literature is befitting, in my estimation.

Response: Thank you for this comment. Accordingly, we have amended the description in the Discussion.

Changes in the reference: See page 10, lines 11–14.

The fourth paragraph discusses pathophysiology and, in its present form, is simplistic and not very well written. This paragraph asks for reformulation followed by careful re-writing.

Response: Thank you for this comment. Accordingly, we have amended the fourth paragraph of the Discussion and the References and have had the text polished by a professional English editing service.

Changes in the text: See page 11, lines 2–20.

Next-to-last paragraph: The seasonal-temperature issue that I have mentioned.

Response: Thank you for this comment. As mentioned above, we have deleted the descriptions and Supplementary Figure 1, which previously showed the seasonal variation of the disease in the manuscript.

Changes in the text: See page 9, line 4 and page 13, line 2.

Last paragraph: There is no mention to the main issue of the study, selection bias -- I stress the point that not only it has to be mentioned but that it must also be discussed -- its sources, how it may affect the study, and so on.

Response: Thank you for this comment. We agree with your point that the study results could have been affected by selection bias owing to the study design. Accordingly, we have amended the limitations section of the Discussion.

Changes in the text: See page 13, lines 3-4.

TABLE 1 --
I know that fL is femtoliters -- but does the reader?
Again, multiple abnormalities point to a common mechanism, and also suggest that the study would benefit from better model construction.

Response: Thank you for this comment, and we apologize for the error in Table 1. Accordingly, we have defined fL in Table 1.

Changes in the text: See Table 1.

Congratulations, this is an interesting paper although the authors just offer preliminary data. I just have a few questions that should be answered:

All the patients with an acute scrotum have a hematologic test at your hospital? Even if the suspected diagnosis is not a testicular torsion?

Response: Thank you for this comment. When patients visit our institution with an acute scrotum requiring emergency surgery, we routinely perform hematologic tests, including CBC examination.

Did you find a cut-off point in the platelet volume to suspect testicular torsion? What about the rest of hematologic parameters?

Response: Thank you for this comment. Within the golden time, the MPV seemed to be the most valuable diagnostic parameter (AUC value: 0.855, cutoff value: 9.35 fL). The AUC value of the rest of the hematologic parameters was lower than that of the MPV within the golden time (WBC count: 0.565; neutrophil count: 0.631; neutrophil ratio: 0.695; NLR: 0.678). The cutoff value of the rest of the hematologic parameters showed low importance and has thus been omitted. The ROC curves of the hematologic parameters (WBC count, neutrophil count, neutrophil ratio, MPV, and NLR) within and after the golden time are shown in Figure 2.