How to Write a Scientific Paper

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Assistant Professor
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Understanding Scientific Writing

- Art

- Learning curve/Experience

- Anyone can write but only well written papers are published in journals with high impact factor
Types of Papers

1. Original Article

2. Review Article

3. Case Studies
Understanding the structure

• Editors:
  ✓ reject ill-prepared manuscripts
  ✓ attempt to improve those accepted

• Referees:
  ✓ provide a detailed criticism of the content of papers submitted

• Authors
The Subject

• Worthy of reporting
• Addition to existing literature
• Do not waste your time on writing a paper that will never be published
Scientific Paper Components

- Title page
- Abstract
- Introduction
- Materials (Patients) and Methods
- Results
- Discussion
- References
- Tables, figures, legends of figures (results) and any acknowledgements
Author’s check list

The author’s check list.

**Introduction**
- Short review
- Shortcomings of the existing reports
- Aim of the study
- Scope of the study

**Patients (or materials) and methods**
- Full description of patients/materials
- Full description of methods
- Study design
- Statistical analysis
- Ethical considerations

**Results**
- Presentation of data
- Correlation of data

**Discussion**
- Introduction to discussion
- Discussion of the results
- Advantages of the study
- Limitations of the study
- Recommendations of authors
Reviewer's check list

The reviewer’s checklist.

Introduction
Are the objectives clear?
Is the importance of the study adequately emphasised?
Is the subject matter of the study new?
Is previous work on the subject adequately cited?

Patients (materials) and methods
Is the study population detailed adequately?
Are the methods described well enough to reproduce the experiment?
Is the study design clear?
Are statistical methods included?
Are ethical considerations provided?

Results
Can the reader assess the results based on the data provided?
Is the information straightforward and not confusing?
Are there adequate controls?
Are statistical methods appropriate?

Discussion
Do the authors comment adequately on all their results?
Have the authors explained why and how their study differs from others already published?
Do the authors discuss the potential problems and limitations with their study?
Are the authors’ conclusions supported by the results?
Title Page

• Title
• List of Authors
• Institutions (affiliations)
• Running Title
• Keywords
• Word Count: limits (< 3000 words)
• Corresponding Author
Prognostic value of cyclooxygenase-2 expression in squamous cell carcinoma of the bladder

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Running title: COX-2 in SCC of the urinary bladder

Key Words: Bladder cancer, biomarkers, Squamous cell carcinoma, COX-2

Word Count: 2803
Abstract Count: 226
References: 39
Figures: 3
Tables: 2

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Title

- Informative
- Specific
- Relatively short: max info in minimum words
- Accurate
- Stimulate the reader to read the rest of the paper
- Try to avoid: conclusion and questions
Title: Examples

• Long-term outcomes of renal tumor radio frequency ablation stratified by tumor diameter: size matters.
• TALL score for prediction of oncological outcomes after radical nephroureterectomy for high-grade upper tract urothelial carcinoma.
• Preoperative multivariable prognostic model for prediction of nonorgan confined urothelial carcinoma of the upper urinary tract.
• Oncological outcomes after radical nephroureterectomy for upper tract urothelial carcinoma: comparison over the three decades.
• Evaluation of vitamin E and selenium supplementation for the prevention of bladder cancer in SWOG coordinated SELECT.
• Utility of biomarkers in the prediction of oncologic outcome after radical cystectomy for squamous cell carcinoma.
• Shock wave lithotripsy versus semirigid ureteroscopy for proximal ureteral calculi (<20 mm): a comparative matched-pair study.
• Clinical outcomes after ureteroscopic lithotripsy in patients who initially presented with urosepsis: matched pair comparison with elective ureteroscopy.
• Residual fragments following ureteroscopic lithotripsy: incidence and predictors on postoperative computerized tomography.
Authors

• Each and Every Author must have been involved in:
  • The conception and design
  • analysis and interpretation of data
  • drafting the article or revising it critically
  • final approval of the version to be published
Author list

• First author: done most of the work
• Equal contribution: shared authorship or advantage to Junior
• Last author: PI/mentor
• Final order of authors list: responsibility of the senior author
Abstract

First text to appear
Should be last to be written

Components:
• Introduction
• Methods
• Results
• Conclusions

Limits:
< 300 or 250 words
Utility of Biomarkers in the Prediction of Oncologic Outcome after Radical Cystectomy for Squamous Cell Carcinoma

Ramy F. Youssef,† Friedrich-Carl von Rundstedt,♦ Payal Kapur, Ahmed Mosbah, Hassan Abol-Enein, Mohamed Ghoneim and Yair Lotan

From the Departments of Urology, University of California-Irvine (RFY), Irvine, California, Urology and Nephrology Center, Mansoura University (RFY, AM, HA-E, MG), Mansoura, Egypt, and Helios Klinikum Wuppertal, University Witten-Herdecke (FCVR), Germany; and Department of Urology, Baylor College of Medicine (FCVR), Houston and Departments of Pathology (RFY) and Urology (FL), University of Texas Southwestern Medical Center, Dallas, Texas.

Purpose: We evaluated the association of multiple biomarkers with clinical outcomes in patients treated with radical cystectomy for squamous cell carcinoma of the bladder to identify the best prognostic panel of markers.

Materials and Methods: Immunohistochemistry for 14 biomarkers was performed on tissue microarray sections of 161 radical cystectomy specimens showing squamous cell carcinoma. Biomarker alterations, pathological features and oncologic outcomes were evaluated. The panel of biomarkers that best predicted the oncologic outcome was determined. Outcomes were stratified based on a prognostic score according to the number of altered biomarkers. The accuracy of oncologic outcome prediction was evaluated by ROC curves.

Results: The study included 151 patients. Pathological stage was T2 in 50%, T3 in 38%, T1 in 6% and T4 in 6% of patients. Median followup was 63.2 months. The best prognostic panel of markers included COX-2, FGF-2, p53, Bax and EGFR. On multivariate Cox regression analysis a prognostic score based on marker alterations was an independent predictor of intermediate and high risk of disease recurrence (HR 3.2, p = 0.008 and HR 15.5, p ≤0.001) and bladder cancer specific mortality (HR 5.2, p = 0.009 and HR 19.4, p ≤0.001, respectively). A multivariate prognostic model incorporating the prognostic score demonstrated significantly better performance to predict the outcome compared to clinicopathological parameters only (0.78 vs 0.64).

Conclusions: Biomarkers have significant potential to predict the outcome of radical cystectomy for squamous cell carcinoma. An increased number of altered markers may identify patients at high risk who might benefit from multimodal treatment approaches.

Key Words: urinary bladder; carcinoma, squamous cell; cystectomy; schistosomiasis; biological markers
Introduction

• Aim and Fundamentals:
  ✓ Short review (background):
    concise, interesting and informative with references (major and recent)
  ✓ Shortcomings of previous studies (convince the reader with importance of your study)
  ✓ Aim of the study (should answer an important question)
  ✓ Scope of the study (a short paragraph or 2 sentences)
• Mission: Logically explain the rationale for the study (why?)
• Importance: Sets the tone of the article, grab reader’s attention
BLADDER cancer is the ninth most common cancer in the world and considerably more common in developing countries.\textsuperscript{1-3} Unlike in UC cases the main SCC risk factors are not environmental exposure such as tobacco but exposure to infectious or inflammatory agents. Although bladder SCC is rare in Western countries, it is more common in a number of developing countries, including Egypt, where schistosomiasis prevails. Prolonged inflammation due to schistosomiasis or chronic indwelling catheters can lead to squamous metaplasia, dysplasia and eventually carcinoma.\textsuperscript{4-6}

Chronic inflammation is associated with multiple cancers, including esophageal, colon, gastric, pancreatic, liver and bladder cancer. Inflammation creates an environment abundant in growth factors and cytokines that favors cell proliferation, migration, angiogenesis and the suppression of apoptosis.\textsuperscript{7} COX-2 is an inducible enzyme responsible for converting arachidonic acid to PG. It is not detectable in most normal tissues but is induced at inflammation sites by cytokines, growth factors and tumor promoters.\textsuperscript{8} COX-2 is over expressed in a range of human malignancies, including bladder SCC.\textsuperscript{9-11} COX-2 over expression is implicated in cellular proliferation, angiogenesis, apoptosis, invasion/motility and immune responses.\textsuperscript{12-16} It is associated with the development and progression of UC\textsuperscript{17-21} as well as SCC.\textsuperscript{9-11} However, there are only a few published studies of COX-2 expression in patients with bladder SCC and they were limited by small patient cohorts. These investigators found COX-2 over expression in bladders with SCC compared to that in normal bladders but they did not evaluate the prognostic significance of COX-2 expression.\textsuperscript{9-11}

We postulated that COX-2 over expression would result in worse outcomes in patients who undergo RC for SCC. Thus, we evaluated the association of COX-2 expression with SCC pathological characteristics and clinical outcomes after RC in a large cohort of patients with long-term followup.
Introduction Evaluation

• Is the aim of the study clear?
• Is the study important? Novel? Adding to existing literature?
• Is previous work adequately cited?
Materials and Methods: *What, Why, & Who*

- **What**: *What* procedures were performed
- **Why**: *Why* were these procedures chosen specifically
- **Who**: *Who* were the test subjects
Materials and Methods

Patients or materials
✓ The patients or materials of the study must be fully described

Methods
✓ Surgical technique, radiological technique, or drugs used
✓ Only new methods need to be described in detail
✓ For a common previously published method, use only a reference
✓ Any manufacturer’s details must be mentioned
Materials and Methods

• Design of the study: prospective vs retrospective, randomized

• Statistical methods: stat test used

• Ethical considerations: informed consent, IRB approved
Prognostic Value of Cyclooxygenase-2 Expression in Squamous Cell Carcinoma of the Bladder

Ramy F. Youssef, Shahrokh F. Shariat, Payal Kapur, Wareef Kabbani, Ahmed Mosbah, Hassan Abol-Enein, Mohamed Ghoniem and Yair Lotan*

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MATERIALS AND METHODS

Patient Population
We reviewed the records and pathological specimens of patients treated with RC and pelvic lymphadenectomy due to bladder cancer in Mansoura, Egypt from 1997 to 2003. After excluding those who received neoadjuvant or adjuvant chemotherapy, or radiotherapy we identified 152 with pure SCC who had sufficient paraffin embedded archival material of the cystectomy specimens available for extensive IHC evaluation. We collected comprehensive clinicopathological data on each patient and entered the data into a database after receiving institutional review board approval.

Patient Followup
All patients were followed for disease progression every 2 months in year 1 and at 6-month intervals thereafter. Generally followup visits consisted of medical history, physical examination and laboratory tests, including serum chemistry evaluation, liver function tests and alkaline phosphatase measurement when clinically indicated.

and/or excretory urography and chest x-ray, were done semiannually or when clinically indicated. Computerized tomography, magnetic resonance imaging and bone scans were performed at treating physician discretion when findings suggested disease progression.

Tissue Microarray Block Construction
Histology, tumor grade and stage were confirmed by blinded review of hematoxylin and eosin stained new sections cut from duplicate archival paraffin blocks in each RC case. Three replicates of 1 mm core diameter samples were collected from a single block in each case and placed on separate, randomly arranged spaces to construct tissue microarray blocks. Sections (5 μm) were obtained from the microarray and stained with hematoxylin and eosin to confirm tumor and finally review tumor histology and other pathological parameters before IHC staining. We used the 2002 UICC TNM classification to pathologically stage the tumors. Tumors were graded from I—well to 3—poorly differentiated.

IHC and Scoring
Immunostaining for COX-2 was done at room temperature on the Dako Autostainer. Reagents were used as supplied in the EnVision™ + System/HRP Labeled Polymer, mouse. Target buffer (ph 6.1) was used for antigen retrieval. Optimum primary antibody dilutions were predetermined for COX-2 (RB-5072-P1 polyclonal rabbit, Thermo Fisher Scientific, Rochester, New York) (dilution 1:200) using known positive control tissues. Appropriate positive and negative controls were used.

We used brightfield microscopy imaging coupled with advanced color detection software with ACIS® III. One of us (PK) blinded to the sample tracking system confirmed all generated scores manually. Cases in which the manual score was discrepant from the score generated by the image analyzer were reviewed by another pathologist (WK) and a consensus score was obtained.

We determined the mean percent of positive cells in 10 random hot spots in each specimen. The mean of the triplicate cores in each case was calculated for data analysis. COX-2 expression was considered normal with no reactivity, or 20% or fewer positive cells and over expressed with greater than 20% positive cells. We assessed the prognostic value of COX-2 using serial increments of cutoffs (range 5% to 90%) with 20% as the best cutoff value to determine bladder cancer outcomes.

Statistical Analysis
We applied Pearson’s chi-square test to examine the relationships of COX-2 expression with pathological parameters. Outcomes were measured out to time of disease recurrence or to bladder cancer specific mortality. The cause of death was determined by treating physicians, chart review and/or death certificates. Univariate recurrence and survival probabilities after RC were estimated using the Kaplan-Meier method and differences were assessed by the log rank statistic. Univariate and multivariate Cox regression models were used to address time to recurrence and cancer specific mortality after RC. In all models proportional hazards assumptions were systematically verified using the Grambsch-Therneau residual based test. All
Radical nephroureterectomy for pathologic T4 upper tract urothelial cancer: can oncologic outcomes be improved with multimodality therapy?


MATERIALS AND METHODS

Patient selection

This was an institutional review board (IRB) approved study, with all participating sites providing the necessary institutional data use agreements prior to initiation of the study. A total of 13 academic centers worldwide provided data. A computerized databank was generated for data transfer. After combining the data sets, reports were generated for each variable to identify data inconsistencies and other data integrity problems. Through regular communication with all sites, resolution of all identified anomalies was achieved before analysis. Prior to the final analysis, the database was frozen and the final data set was produced for the current analysis. This study comprised 1464 patients who underwent RNU at 13 centers between 1987 and 2007. 69 patients with pathologically confirmed T4 at RNU were identified and formed the cohort of interest for this study. The choice to perform LND was determined by the surgeon and the standardization of LND was impossible due to the multicenter and retrospective design of the study.

Pathologic Evaluation

All surgical specimens were processed according to standard pathologic procedures, and all slides were re-reviewed by genitourinary pathologists according to prospectively defined uniform criteria. All pathologists were blinded to clinical outcomes. Tumors were staged according to the 7th edition of the American Joint Committee on Cancer-Union International Contre le Cancer (AJCC-UICC) Tumor-Node-Metastasis (TNM) classification [5]. Tumor grades were assessed according to the 1998 WHO/ISUP (International Society of Urologic Pathology) consensus classification [6]. In addition, all UTUCs were evaluated for tumor location (renal pelvis vs. ureter), pattern of tumor growth (papillary vs. sessile), presence of lymphovascular invasion (LVI), and concomitant CIS in the nephroureterectomy specimen.

Surveillance Regimen

Follow-up was performed according to institutional protocols. Patients were generally followed every three months for the first year following RNU and every six months from the second year. Follow-up consisted of a history, physical examination, routine blood work and serum chemistry studies, urinary cytology, chest radiography, cystoscopy, and radiographic evaluation of the contralateral upper urinary tract. Elective bone scans, chest computerized tomography (CT), or magnetic resonance imaging were performed when clinically indicated.

Outcome Evaluation and Statistical Analysis

Disease free survival (DFS) and cancer specific survival (CSS) were estimated using the Kaplan-Meier survival analysis. Disease recurrence was defined as local failure in the nephroureterectomy bed, regional lymph nodes (LN), or distant metastasis after RNU. The period of DFS was defined as the time between the date of RNU and the development of local recurrence or distant me-
Methods Evaluation

• What is kind of the study? Study design? Study population?
• Are methods reproducible?
• Are statistical methods included and sound?
• Are ethical considerations provided, if necessary?
Results

Presentation of data with stat analysis

☑ Text: organized, concise, not repeated in tables or figures

☑ Figures or Graphs: simple, clear, scientifically attractive

☑ Tables
A good table:

- single unit of communication
- supply maximum information with minimum words
- not present in the text, to avoid redundancy

- Each table with number and title
- Make sure number of table appears correct in the text
RESULTS

Patient Demographics and Clinicopathological Findings

Of the 152 study patients 99 were men and 53 were women. Mean age at diagnosis was 51.9 years (median 51, range 36 to 74), including 52.8 (median 53, range 38 to 74) and 51 years (median 50.5, range 36 to 66) in those with over expressed and normal COX-2, respectively (p = 0.22). Presenting stage was T2 or greater in 140 patients (93.4%) and only 6.6% presented with T1 tumors. Grade was 1 to 3 in 53.3%, 40.1% and 6.6% of cases, respectively. Gross and cystoscopic descriptions were available for all tumors. Nodular or fungating tumors were seen in 139 cases (91.5%). The remaining tumors were ulcerating (4 cases), fibrillar (3) or papillary (3), or showed another configuration (3). Tumor involved a single site in 132 cases, was extensive to involve multiple vesical walls in 10 and was multifocal in another 10. Commonly involved regions were the lateral walls in 59 patients, posterior wall in 34, anterior wall in 23 and dome in 15. Bilharzial infection was found in 123 patients (80.9%), LN invasion was found in 46 (30.3%) and LVI was found in 24 (15.8%). An average of 22 LNs (maximum 70) was removed during RC with an average of 1 positive LN (maximum 30). Positive surgical margins were found in 5 cases (3%). The table lists patient characteristics and the association of COX-2 expression with clinicopathological parameters. Median follow-up was 63.2 months (range 0 to 100).

Association of COX-2 Over Expression

Pathological characteristics. Figure 1 shows representative IHC staining results for COX-2 in bladder SCC cases. COX-2 expression was homogenous with predominant expression localized to the cell cytoplasm. Overall 74 study patients (48.7%) showed COX-2 over expression. The table shows the association between COX-2 over expression and clinicopathological parameters. COX-2 over expression was associated with pathological T stage (p = 0.003) and grade (p = 0.049).

Clinical outcomes (recurrence and survival). Mean ± SD 5-year disease-free and cancer specific survival rates in the 152 study patients were 67% ± 4% and 78% ± 4%, respectively. Kaplan-Meier analyses revealed that COX-2 over expression was associated with an increased probability of tumor recurrence and bladder cancer specific mortality (p = 0.03 and 0.02, respectively, fig. 2). On multivariate Cox proportional hazards regression analysis adjusted for the effects of

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Descriptive characteristics in patients with SCC and association of COX-2 expression with clinicopathological parameters

<table>
<thead>
<tr>
<th>No. Pts (%)</th>
<th>No. Over Expressed COX-2 (%)</th>
<th>No. Normal COX-2 (%)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total</td>
<td>152</td>
<td>74 (48.7)</td>
<td>78 (51.3)</td>
</tr>
<tr>
<td>Gender:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>53 (34.9)</td>
<td>26 (49.1)</td>
<td>27 (50.9)</td>
</tr>
<tr>
<td>M</td>
<td>99 (65.1)</td>
<td>48 (48.5)</td>
<td>51 (51.5)</td>
</tr>
<tr>
<td>Pathological stage:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pT1</td>
<td>10 (8.8)</td>
<td>3 (30)</td>
<td>7 (70)</td>
</tr>
<tr>
<td>pT1a</td>
<td>24 (15.8)</td>
<td>5 (20.8)</td>
<td>19 (79.2)</td>
</tr>
<tr>
<td>pT1b</td>
<td>52 (34.2)</td>
<td>29 (55.6)</td>
<td>23 (44.2)</td>
</tr>
<tr>
<td>pT3</td>
<td>57 (37.5)</td>
<td>35 (61.4)</td>
<td>22 (38.6)</td>
</tr>
<tr>
<td>pT4</td>
<td>9 (5.9)</td>
<td>2 (22.2)</td>
<td>7 (77.8)</td>
</tr>
<tr>
<td>Tumor grade:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>81 (53.3)</td>
<td>32 (39.5)</td>
<td>49 (60.5)</td>
</tr>
<tr>
<td>II</td>
<td>61 (40.1)</td>
<td>36 (59)</td>
<td>25 (41)</td>
</tr>
<tr>
<td>III</td>
<td>10 (6.6)</td>
<td>6 (60)</td>
<td>4 (40)</td>
</tr>
<tr>
<td>pN stage:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>106 (69.7)</td>
<td>54 (50.9)</td>
<td>52 (49.1)</td>
</tr>
<tr>
<td>1–3</td>
<td>46 (30.3)</td>
<td>20 (43.5)</td>
<td>26 (56.5)</td>
</tr>
<tr>
<td>Bilharziasis:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absent</td>
<td>29 (19.1)</td>
<td>14 (48.3)</td>
<td>15 (51.7)</td>
</tr>
<tr>
<td>Present</td>
<td>123 (80.9)</td>
<td>60 (48.8)</td>
<td>63 (51.2)</td>
</tr>
<tr>
<td>DNA ploidy:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diploid</td>
<td>83 (54.8)</td>
<td>37 (44.6)</td>
<td>46 (55.4)</td>
</tr>
<tr>
<td>Tetráploid</td>
<td>28 (18.4)</td>
<td>13 (46.4)</td>
<td>25 (53.6)</td>
</tr>
<tr>
<td>Aneuploid</td>
<td>41 (27)</td>
<td>24 (58.5)</td>
<td>27 (41.5)</td>
</tr>
<tr>
<td>LVI:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absent</td>
<td>128 (81.2)</td>
<td>62 (48.4)</td>
<td>66 (51.5)</td>
</tr>
<tr>
<td>Present</td>
<td>24 (15.8)</td>
<td>12 (50)</td>
<td>12 (50)</td>
</tr>
<tr>
<td>Concurrent Ca in situ:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absent</td>
<td>141 (94.1)</td>
<td>71 (49.7)</td>
<td>72 (50.3)</td>
</tr>
<tr>
<td>Present</td>
<td>9 (5.9)</td>
<td>3 (33.3)</td>
<td>6 (66.7)</td>
</tr>
</tbody>
</table>
Figure 1. Bladder SCC COX-2 expression. A, stronger expression with higher grade. B, stronger expression with higher stage. Reduced from ×100.
Figure 2. Kaplan-Meier curves reveal association of COX-2 expression and patient outcome. A, recurrence-free survival. B, cancer specific survival.
Results Evaluation

✓ Straightforward or confusing?
✓ Are the statistical methods appropriate?
✓ Can you draw any clear conclusions based on results?
Discussion

1. Start with the most important finding and main results of your study
2. Inverted Pyramid: most important to less important results
3. Comparison of findings from current study with previous studies
4. Similarities and differences to previous studies
5. Possible explanation(s) for the different findings if applicable
6. Advantages and limitations
7. Recommendations, take home message and proposal for future research
References

• Each journal has its own style of references
• Read instruction and examine recent copies of the journal
• All references shall be written in same style with same arrangement
• Recent references are better than older references
• Styles of references can vary

➢ **Vancouver system** = Commonly used in medical journals
  - References are arranged numerically according to their order of their appearance in the text

➢ **Harvard system** = Commonly used in a thesis
  - References are arranged alphabetically written as the name of the author(s) followed by the year of publication
References

• Learn Endnote:
  ✓ automatically arrange references for you
  ✓ share your library with coauthors and mentors

• Learn Pubmed
  ✓ Search literature
  ✓ Read
  ✓ Collect data
Writing order

• Methods and results
• Introduction and discussion
• Abstract and conclusions
• Review
• Finalize title
• Review again, edit and improve
• Review again, edit and improve after revisions and comments by your mentor and coauthors
• Submit and get ready to start another journey after submission
Common Reasons for Rejection

• Lack of relevance to journal
• Not styled correctly to journal’s standards
• Poorly designed study
• Poorly written
• Conclusions stated were unjustified
• Reviewer/editor bias