

Neutrophil to lymphocyte ratio and muscular invasion in early-stage bladder cancer: a meta-analysis

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Summary. *Background:* Bladder cancer is the ninth most common cancer worldwide. In its early stages, invasion of the muscle layer of the bladder is the major determinant for cystectomy. The aim of this meta-analysis was to evaluate the potential role of the neutrophil to lymphocyte ratio (NLR) in predicting muscular invasion in early-stage bladder cancer. *Methods:* A systematic literature search was conducted in Medline, PubMed, Scopus and Clinicaltrials.gov databases for English language articles published in the last decade. *Results:* Five studies fulfilling the eligibility criteria were identified (1,612 participants, 1,217 with NMIBC and 395 with MIBC). Pooled results showed that NLR values were significantly higher in patients with MIBC (SMD: 0.45, 95% CI: 0.18-0.73; p=0.001). *Conclusions:* NLR is significantly higher in patients with MIBC in comparison to those with NMIBC. This simple, widely available and relatively inexpensive parameter might be useful for risk stratification in patients with early-stage BC.

Key words: cancer, bladder, urothelial, muscle invasion, NLR

Introduction

Bladder cancer (BC) is the ninth most common cancer worldwide, with approximately 430,000 new cases estimated in 2012 (1). Its incidence and prevalence peak in the seventh to eighth decade of life, and it is 3 to 4 times more common in men than in women. Incidence rates are highest in Europe, the United States, and Egypt (2). In the United States, the incidence and mortality rates have been stable during the past three decades. In 2012, an excess of 165,000 BC-related deaths were estimated worldwide (1). Urothelial cancer is the most common cancer of the bladder, accounting for more than 95% of bladder cancers in several populations (3, 4).

From a clinical perspective, BC is classified as non-muscle-invasive bladder cancer (NMIBC, 70%)

and muscle-invasive bladder cancer (MIBC, 30%), because invasion of the muscle layer is the major determinant for performing a cystectomy (5). The pathological confirmation of muscular invasion derives from biopsies obtained using a transurethral approach (TUR). However, this approach has some limitations, especially in cases of repeated resections (6,7). For this reason, the identification of biomarkers associated with muscular invasion in early-stage BC might be particularly useful for risk stratification and management.

In recent years, the neutrophil to lymphocyte ratio (NLR), a simple, easy to perform, and relatively inexpensive parameter has attracted particular interest, because it accurately reflects systemic inflammatory alterations in numerous diseases, including cancer (8-12). Several authors have investigated the prognostic role of the NLR in bladder cancer (13-15). A number

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of studies have also been performed to investigate differences in NLR values between BC patients with and without muscular invasion. We conducted a systematic review and meta-analysis of the studies investigating the associations between the NLR and muscular invasion in early-stage BC.

Materials and Methods

Eligibility criteria

Human studies were considered eligible if they met the following criteria: (1) pre-treatment assessment of blood NLR, (2) compared subjects with NIMBC and IMBC, (3) BC diagnosed histologically in accordance with the WHO classification system (16) and staged in accordance with the American Joint Committee on Cancer (AJCC) staging system (17), (4) English language, (5) full-text publications, (6) basic demographic and clinical data available.

Search strategy and study selection

A systematic literature search was conducted in Medline, Pubmed, Scopus and Clinicaltrials.gov databases for English language articles published between 2007 and 2017, using the following terms: “neutrophil to lymphocyte ratio” OR “NLR” AND “bladder cancer” OR “urothelial cancer”.

Abstracts were independently screened by two investigators. If relevant, full articles were retrieved. References in these articles, citing relevant reviews or original studies were also accessed to identify additional eligible studies. Any disagreement between the reviewers was resolved by a third investigator. We used the Newcastle-Ottawa Scale (NOS) to assess the quality of each study (19). The Newcastle-Ottawa scale evaluated three components: selection of the cohort, comparability of cohorts on the basis of the design or analysis, and assessment of exposure and outcomes of interest. Studies achieving six or more stars were considered to be of high quality.

Statistical analysis

Standardized mean differences (SMD), and 95% confidence intervals (CIs), were used to construct forest plots of continuous data and to evaluate differences in NLR values between patients with NIMBC and patients with IMBC. $P < 0.05$ was considered statistically significant. In one study (19) the standard deviation (SD) was estimated from the median and range.

Heterogeneity of SMD across studies was tested by using the Q statistic (significance level at $p < 0.10$). The I^2 statistic, a quantitative measure of inconsistency across studies, was also calculated ($I^2 < 25\%$, no heterogeneity; I^2 between 25% and 50%, moderate heterogeneity; I^2 between 50% and 75%, large heterogeneity; and $I^2 > 75\%$, extreme heterogeneity) (20, 21). Due to the high heterogeneity, a random-effects model was used to calculate the pooled SMD and corresponding 95% confidence intervals.

To evaluate the presence of potential publication bias, the association between study size and magnitude of effect were analysed by means of Begg's adjusted rank correlation test and Egger's regression asymmetry test at the $p < 0.05$ level of significance (22). Sensitivity analysis was conducted to investigate the influence of an individual study on the overall risk estimate by sequentially excluding one study in each step (23). Statistical analyses were performed using MedCalc for Windows, version 15.4 64 bit (MedCalc Software, Ostend, Belgium) and Stata 14 (STATA Corp., College Station, TX, USA). Reporting methods comply with the PRISMA statement.

Results

A flow chart describing the study selection is presented in Figure 1. We initially retrieved 281 studies. Of these, 132 duplicates were excluded after an initial screening, and 126 further studies were subsequently excluded after abstract evaluation. After full-text review of the remaining 23 articles, 18 studies were excluded because they did not meet the inclusion criteria. Five studies were included in the final meta-analysis (5, 19, 24-26).

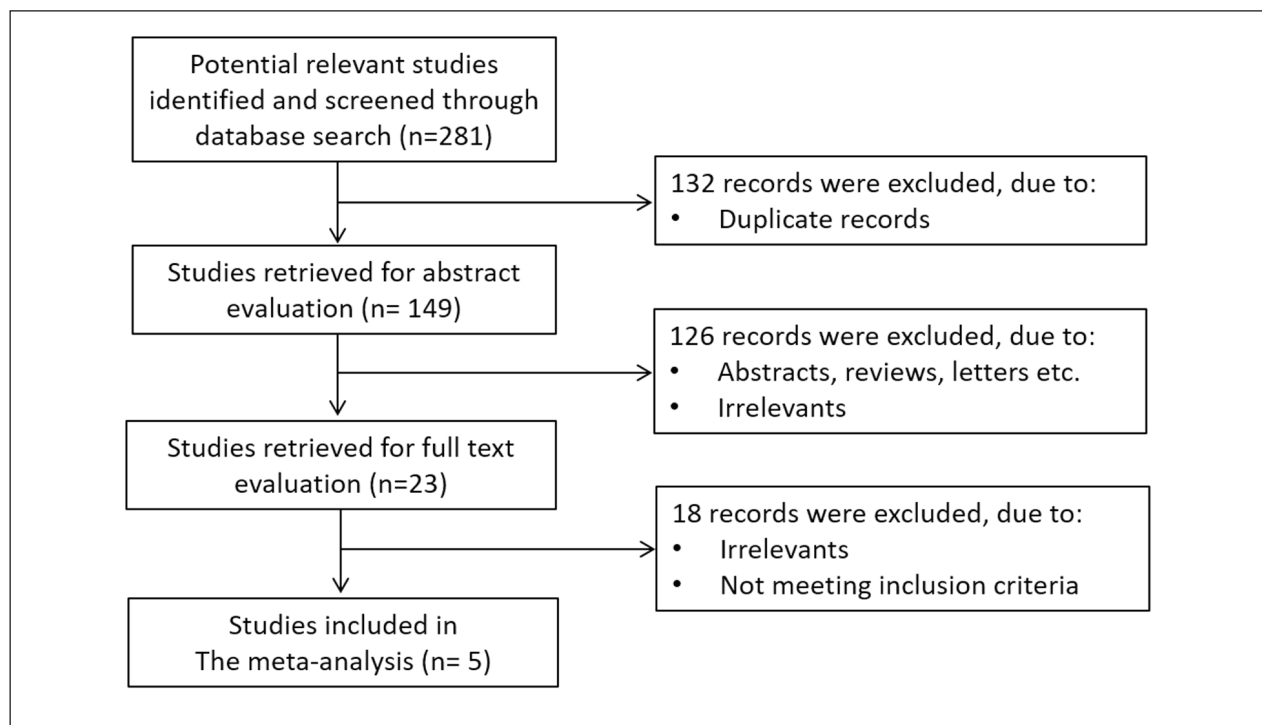


Figure 1. Flow chart showing the study search and selection procedure

Table 1. Summary of the studies on NMIBC vs MIBC included in the meta-analysis

First Author, Year, Country	Ceylan et al. 2014, Turkey	Kaynar et al. 2014, Turkey	Celik et al. 2016, Turkey	Ma et al. 2016, China	Tazeh et al. 2017, USA
Study design	R	R	R	R	R
NOS					
Patients enrolled (total n)	198	291	222	669	232
NMIBC					
n	162	192	162	579	122 (Ta)
Age (mean \pm SD, years)	63 \pm 11.1	64 \pm 13.0	71.8 \pm 10.9	65.2 \pm 11.9	NA
Gender (M/F)	150/12	156/36	141/21	NA	NA
NLR Mean \pm SD	3.36 \pm 2.9	2.4 \pm 0.1	3.44 \pm 2.0	2.71 \pm 2.5	2.2 \pm 1.0
MIBC					
n	36	99	60	90	110 (T2)
Age (mean \pm SD, years)	72.6 \pm 10.3	75 \pm 10.0	75.7 \pm 10.2	67 \pm 11.4	NA
Gender (M/F)	32/4	85/14	51/9	NA	NA
NLR Mean \pm SD	4.14 \pm 2.8	2.9 \pm 0.2	4.65 \pm 2.8	4.66 \pm 8.0	3.4 \pm 1.8

R: retrospective; NA: not available

The characteristics of the five studies investigating the NLR in NMIBC vs MIBC, and their NOS evaluation, are presented in Table 1. All studies were retrospective and comprised a total of 1,612 patients, 1,217 with NMIBC and 395 with MIBC. The male/female ratio in the NMIBC and MIBC groups was not available in all studies.

The forest plot showed higher mean NLR values in MIBC patients, when compared to those with NMIBC, in all the studies (Figure 2). Because of the substantial heterogeneity between studies ($I^2=80.1\%$, $p < 0.001$), random-effects models were used. Pooled results showed that NLR values were significantly higher in patients with MIBC (SMD: 0.45, 95% CI: 0.18–0.73; $p=0.001$). No evidence of publication bias was noted (Begg, $p=0.81$; Egger, $p=0.98$). Results stability was evaluated through sensitivity analysis (Figure 3). The corresponding pooled SMD values were not substantially altered when single studies were removed, suggesting that the results of the meta-analysis were stable.

Discussion

Although the presence of leukocytes in tumour tissues was known since the 19th century, the evidence that inflammation plays a critical role in the genesis and progression of human cancers has only been generated during the last decade (27). The NLR is a simple index obtained from complete blood counts, which reflects the systemic inflammatory status of the organism, on the basis of disease-related modifications of the most representative cell populations of inflammation. It has been recently proposed as a predictor of the onset, progression, and prognosis of several chronic inflammatory diseases and cancers (8–15). Wei et al. performed a meta-analysis to investigate the prognostic role of NLR in urinary cancers, including renal cell, upper tract urothelial, prostate and bladder cancers (13). The authors found that elevated NLR was a poor predictor for survival in patients with urinary cancers. Similar results were reported in other meta-analyses

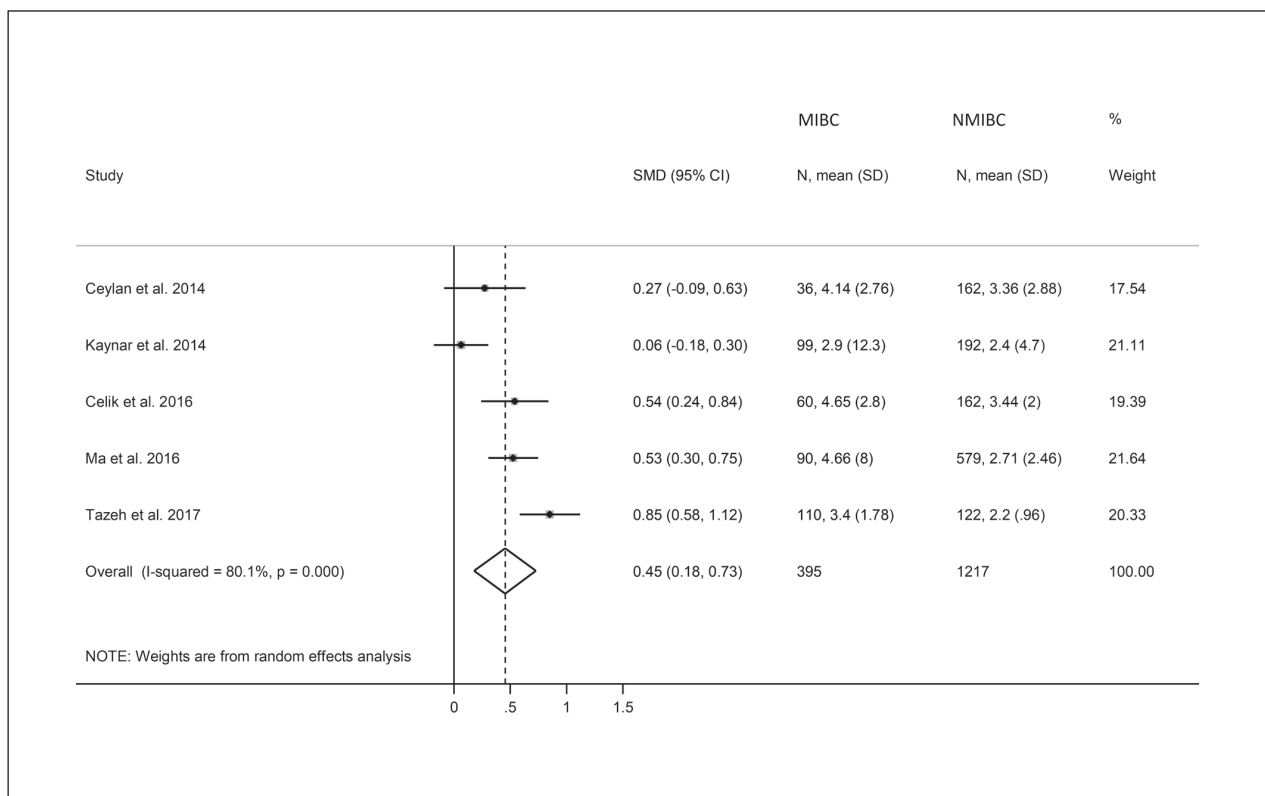


Figure 2. Forest plot of studies examining NLR and muscular invasiveness of early bladder cancer

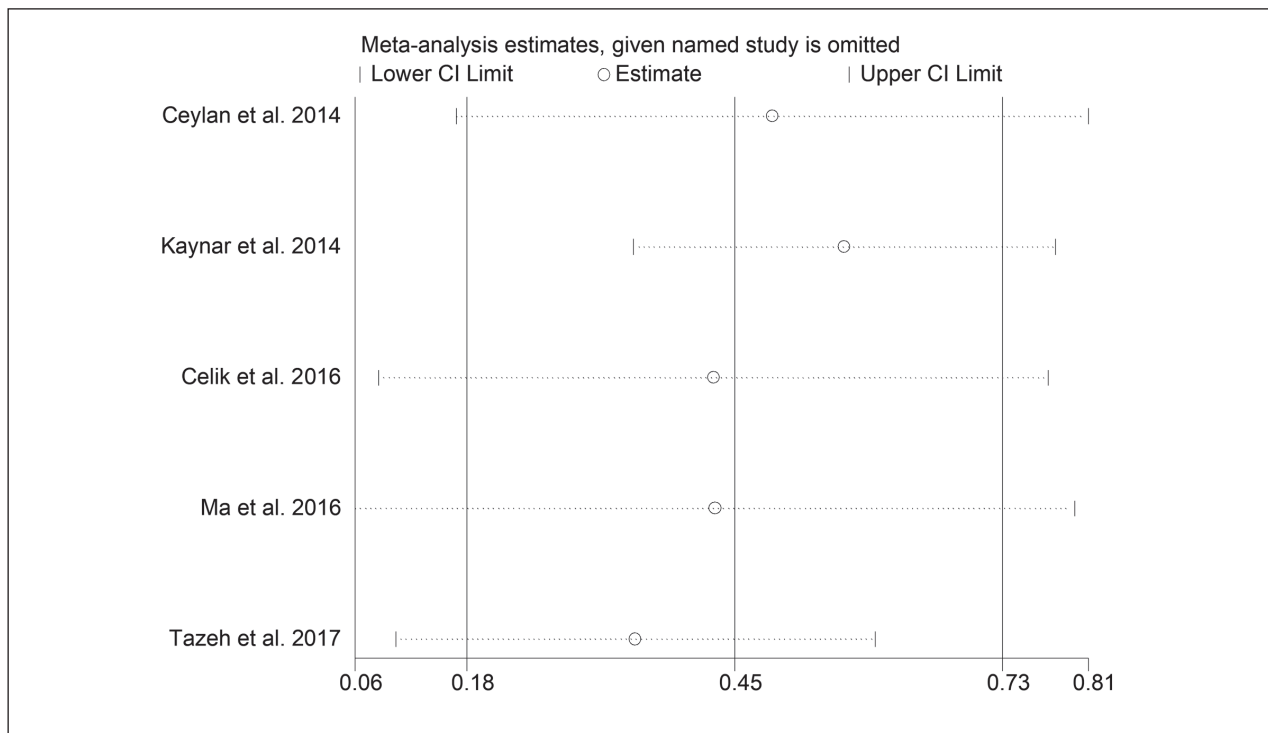


Figure 3. Sensitivity analysis of the association between NLR and muscular invasiveness of early bladder cancer

which analysed the prognostic roles of NLR in upper tract and urothelial BC (28) or urothelial BC alone (14, 29). These findings confirmed the potential prognostic roles of NLR in bladder cancer.

In this meta-analysis, we focused on a different potential clinical use of the NLR in early-stage BC, by comparing values between patients with and without muscular invasion. In five retrospective studies with adequate clinical, pathological and laboratory data, the NLR values were significantly higher in patients with MIBC (SMD: 0.45, 95% CI: 0.18-0.73; $p=0.001$) in comparison to those with NMIBC. This is likely to reflect the greater systemic inflammatory state in patients with muscular invasion, and thus, higher stage BC. Numerous previous studies have demonstrated the positive correlation between NLR levels and higher neoplastic stage in urinary and other malignancies (28, 30-32).

No consistent biases were detected by means of the Begg and Egger tests, however this may be influenced by the relatively low number of articles identified. Moreover, other limitations should be acknowledged,

including the small number of studies identified and their retrospective design, which furthermore preclude meta-regression analyses. On the other hand, this is the first meta-analysis that evidences a potential role of NLR as a predictor of muscular invasion in early bladder cancer. The availability of such a predictor may be useful in clinical practice for the assessment of muscular invasion, especially in cases in which the pathological examination may be inconclusive. An additional advantage is the fact that NLR is a simple, low-cost, widely available index, which can be easily calculated from complete blood count tests. Nevertheless, further well-designed clinical trials are warranted to establish its potential clinical usefulness and applications.

Conclusions

The present meta-analysis, the first to evaluate the potential role of NLR in detecting muscular invasion of early stage BCs, showed that the NLR is consistently higher in patients with MIBC in com-

parison to those with NMIBC. Therefore, the NLR, a simple, widely available, and relatively inexpensive marker, may be useful in risk stratification and staging of patients with early-stage BC, especially those with a challenging pathological confirmation. Further prospective studies are required to confirm these findings.

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