Postdiagnosis aspirin use and overall survival in patients with melanoma

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Background: Mouse studies show that tumor-derived prostaglandins and platelets promote melanoma progression and immune evasion.

Objective: Determine whether aspirin confers longer survival in patients with melanoma.

Methods: A retrospective cohort study of 1522 patients at Indiana University Health who had melanoma diagnosed between 2000 and 2014 and were followed up through September 2016.

Results: Aspirin use was associated with longer overall survival in univariate analysis and after controlling for age, sex, stage, and treatment modalities (hazard ratio [HR], 0.58; 95% confidence interval [CI], 0.45-0.75). Aspirin use was not associated with survival in patients with in situ and stage I melanoma but was associated with better survival in stages II (HR, 0.45; 95% CI, 0.24-0.82) and III (HR, 0.57; 95% CI; 0.34-0.96). No statistical significance was observed in stage IV patients (HR, 0.55; 95% CI, 0.27-1.13). In turn, melanoma in patients using aspirin before diagnosis was less likely to be diagnosed in stages III or IV.

Limitations: Observational study.

Conclusions: Aspirin could provide a survival advantage in melanoma. Clinical trials investigating the therapeutic potential of aspirin are warranted. (J Am Acad Dermatol https://doi.org/10.1016/j.jaad.2017.12.076.)

Key words: aspirin; melanoma; platelets; stage; survival; treatment.

Cutaneous melanoma is the fifth most common cancer in the United States, with 87,110 new invasive melanoma cases and 9730 deaths estimated in 2017.1 The 5-year overall survival rate is 18% in patients with distant metastasis.4 New treatments of advanced melanoma such as B-Raf proto-oncogene inhibitors, serine/threonine kinase inhibitors2 and antibodies targeting programmed cell death 1 and cytotoxic T-lymphocyte associated protein 43,4 have led to markedly improved outcomes compared with those of previously used chemotherapies. However, the outcomes are still unsatisfactory and new treatments are under investigation.

Abbreviations used:
- CPH: Cox proportional hazards
- CI: confidence interval
- HR: hazard ratio
- TGFβ: transforming growth factor-β

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We recently showed that platelets enhance melanoma growth by suppressing antitumor T-cell immunity in murine models, and antiplatelet medications enhance the efficacy of melanoma immunotherapy.\(^5\) Moreover, melanoma-derived prostaglandins weaken antitumor immunity, and blocking the cyclooxygenase pathway with aspirin improves the efficacy of immunotherapy with anti–programmed cell death 1 antibodies in mice.\(^6\) Thus, aspirin can potentially improve melanoma outcomes by inhibiting platelets and tumor-derived prostaglandin production.

Clinically, cancer-associated thrombocytosis correlates with poor clinical outcomes in a number of cancers.\(^7-9\) In long-term follow-up of patients in cardiovascular clinical trials, aspirin use correlated with lower incidence and mortality from colorectal cancer,\(^10\) and use of aspirin after diagnosis was associated with improved survival.\(^11\) Case-control studies also pointed to a possible protective role of aspirin against esophageal, gastric, biliary, and breast cancers.\(^12\) We have also shown that postdiagnosis use of aspirin is associated with better overall survival in head and neck cancer.\(^9\) Importantly, several studies point to a negative association between aspirin use and incidence of melanoma, although the evidence for that has been inconsistent.\(^13,14\) Aspirin use before diagnosis was also associated with smaller Breslow thickness, which is 1 of the strongest prognostic predictors in melanoma.\(^15\) However, the potential benefit from postdiagnosis aspirin use in melanoma is not studied, and its potential effect in different stages of cancer is not well understood either.

Given the epidemiologic evidence of the protective role of aspirin in some cancers, along with the role of platelets and tumor-derived prostaglandins in shaping cancer behavior, this study investigated aspirin as a protective factor in patients with melanoma. In light of the known contribution of platelets to metastasis,\(^16,17\) we also analyzed the relation between prediagnosis aspirin use and melanoma stage at time of diagnosis.

**METHODS**

**Study design**

This study was approved by the institutional review board at Indiana University Health. Patient data were collected from the Simon Comprehensive Cancer Center registry at Indiana University Health, which obtains information by using pathology reports, electronic medical records, radiation and chemotherapy oncology reports, and coding and billing data. This study included patients in whom melanoma was diagnosed between January 1, 2000, and December 31, 2014, with follow-up through September 28, 2016. Data included age, sex, stage, primary tumor site, treatment modalities, tumor characteristics, and patient outcomes. The registry included 2452 melanoma cases. Because the vast majority of patients were white (98%), the main analysis was limited to this patient population (n = 47 excluded). Sensitivity analysis including nonwhite patients was also performed (Supplemental Table I; available at http://www.jaad.org). Patients with more than 1 primary melanoma at date of diagnosis were also excluded (n = 93), as were those who used aspirin only before diagnosis (n = 86). We also excluded patients who lacked complete TNM staging in the registry (n = 704). This resulted in a total of 1522 patients for survival analysis. Overall survival was calculated as the interval from the time of diagnosis to death. The survival times for patients alive at the end of the study period were censored. Patients lost to follow-up in this time period were censored at the date of last contact.

For each patient, further information was obtained from the Indiana University Health Clinical Data Warehouse. This included intake of aspirin and corresponding dates based on prescriptions and/or documentation of use in the medical records and medical comorbidities based on International Classification of Diseases, Ninth Revision and International Classification of Diseases, Tenth Revision codes.

**Data analysis**

Descriptive statistics were used to summarize continuous and discrete variables. Continuous variables were compared by using the 2-sided \(t\) test and discrete variables were compared by using the chi-square or 2-sided Fisher’s exact test, depending on the pertinent sample size. To determine whether aspirin confers better overall survival, the primary end point was defined as death from any cause.
Kaplan-Meier methods were used to estimate the survival probabilities and corresponding 95% confidence intervals (CIs). Survival curves were compared by using log-rank tests. We used the likelihood ratio test to assess the statistical significance of the interaction between aspirin use and stage. The association between aspirin use and survival was evaluated by fitting Cox proportional hazards (CPH) regression models. We first fit univariate CPH models to investigate the unadjusted associations between aspirin use and survival. We then fit multivariable CPH models adjusting for significant baseline covariates \((P \leq .05)\), including age (continuous), sex, treatment, and stage (model 1). In addition to the variables in model 1, cardiovascular comorbidities at time of diagnosis were also adjusted for (model 2).

Because of the significant interaction between stage and aspirin use on survival (likelihood ratio test, \(P < .01\)), we also fit stratified CPH regression models for the 5 stage groups (in situ and stages I, II, III, and IV). The associations were summarized by using hazard ratios (HRs) and 95% CIs. The CPH assumption was assessed visually and with the Schoenfeld residuals test for each stage-specific model. The proportional hazards assumption was met in each model \((P > .05)\). Given that immunotherapy is currently 1 of the mainstay treatments for patients with unresectable stage III and stage IV melanoma, we also assessed the interaction between aspirin use and immunotherapy on survival. Finally, postdiagnosis aspirin users were stratified into those who also took aspirin before diagnosis and those who did not to evaluate the impact of prediagnosis aspirin use on survival.

The association between prediagnostic aspirin use and melanoma clinical and pathologic features was assessed by using logistic regression analysis to generate the relative risks (RRs) and 95% CIs. Age and sex were then adjusted for in the multivariable analysis model. Melanoma-specific features included Breslow thickness, mitotic count, ulceration, primary tumor regression, lymphovascular invasion, and stage. The RRs for melanoma being diagnosed in stages III and IV in aspirin users versus nonusers were calculated by using the in situ stage as a reference.
reference. All statistical analyses were done with Stata software (version 13.0, StataCorp LP, College Station, TX).

results

Patient characteristics at baseline
This cohort was roughly balanced in sex distribution, with slightly more males than females and an association between male sex and aspirin use (Table I). Aspirin users were older than nonusers and were more likely to have hypertension, diabetes mellitus, dyslipidemia, and cardiovascular diseases (cerebrovascular, ischemic heart, and other atherosclerotic diseases) at diagnosis. Treatment modalities were mostly comparable between aspirin users and nonusers. Postdiagnosis aspirin use was also associated with earlier stage of disease, where melanoma was diagnosed in stages III and IV in fewer patients among aspirin users. Anatomic site was marginally different between the 2 groups; with fewer unspecified sites in aspirin users (2.3%) than in nonusers (7.2%).

Postdiagnosis aspirin use and overall survival
Upon univariate analysis, aspirin use after diagnosis was associated with longer overall survival (HR, 0.66; 95% CI, 0.51-0.85; \( P = .001 \)) (Tables II and III and Fig 1). This trend persisted after adjusting for age, sex, stage, and treatment modalities (HR, 0.58; 95% CI, 0.45-0.75; \( P = .001 \)) (Table II). Adjusting for cardiovascular diseases and comorbid risk factors at diagnosis had no appreciable impact on the association between aspirin use and survival (Table II). Sensitivity analysis including nonwhite patients yielded similar findings (Supplemental Table I).

We observed a statistical interaction between aspirin use and clinical stage on survival. In our stage-specific analyses, there was no significant association between aspirin use and survival in early-stage melanoma: in situ (HR, 0.85; 95% CI, 0.45-1.61; \( P = .61 \)) and stage I (HR, 0.96; 95% CI, 0.54-1.71; \( P = .89 \)). However, there was an inverse association between aspirin use and mortality in stage II (HR, 0.45; 95% CI, 0.24-0.82; \( P = .009 \)) and stage III (HR, 0.57; 95% CI, 0.34-0.96; \( P = .03 \)). No statistical significance was observed in stage IV (HR, 0.55; 95% CI, 0.27-1.13; \( P = .10 \)) (Table II).

Patients who used aspirin after diagnosis experienced longer overall survival regardless of whether they used aspirin before diagnosis (Table II). In addition, stage III and IV patients receiving immunotherapy (checkpoint blockers, interferons, and/or interleukin 2) displayed an association between aspirin use and survival similar to that in the rest of the patients in these stages (no interaction between aspirin use and immunotherapy on survival [\( P = .51 \)]).

<p>|</p>
<table>
<thead>
<tr>
<th>Table II. Postdiagnosis aspirin use and overall survival</th>
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<tr>
<td>Variables</td>
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<td>--------------------------------------------------------</td>
</tr>
<tr>
<td>HR\textsubscript{unadjusted} (95% CI)</td>
</tr>
<tr>
<td>HR\textsubscript{adjusted 1} (95% CI)</td>
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<tr>
<td>HR\textsubscript{adjusted 2} (95% CI)</td>
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<tr>
<td>HR\textsubscript{adjusted} stratified by stage</td>
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<tr>
<td>In situ (n = 421)</td>
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<tr>
<td>Stage I (n = 451)</td>
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<tr>
<td>Stage II (n = 221)</td>
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<tr>
<td>Stage III (n = 304)</td>
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<tr>
<td>Stage IV (n = 125)</td>
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<tr>
<td>HR\textsubscript{adjusted} stratified by prediagnosis aspirin use</td>
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<tr>
<td>Aspirin before and after diagnosis</td>
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<td>Aspirin after diagnosis only</td>
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CI, Confidence interval; HR, hazard ratio.

1\textsuperscript{*}HR adjusted for age, sex, stage, and treatment modality.

2\textsuperscript{*}Adjusted for hypertension, dyslipidemia, diabetes mellitus, and cardiovascular diseases present at diagnosis in addition to the variables in HR\textsubscript{adjusted 1}.

3\textsuperscript{*}Adjusted for age, sex, and treatment.

Prediagnosis aspirin use is associated with earlier stage of melanoma
Next, the association between prediagnosis aspirin use and tumor characteristics was examined. Prediagnosis aspirin use corresponded to any prescription or patient-reported use before the date of diagnosis. In univariate analysis, patients who used aspirin before diagnosis were less likely to have melanoma diagnosed in stages III and IV (Table IV). Stage III melanoma was present in 20.9% of nonusers versus in 11.7% of users (RR\textsubscript{univariate}, 0.28; 95% CI, 0.13-0.59; \( P = .001 \)). This association persisted after
controlling for age and sex (RRMultivariable, 0.33; 95% CI, 0.15-0.73; \( P = .019 \)). Similarly, stage IV disease was observed in 9.9% of nonusers and 2.6% of aspirin users (RRUnivariate, 0.13; 95% CI, 0.03-0.55; \( P = .006 \) and RRMultivariable, 0.13; 95% CI, 0.03-0.57; \( P = .007 \)). Concordantly, melanoma in situ was diagnosed in 25.3% of nonusers compared with 50.7% of aspirin users.

On the other hand, prediagnosis aspirin use was not associated with Breslow thickness, mitotic count, primary tumor regression, or lymphovascular invasion. However, aspirin was associated with increased risk for ulceration, with a prevalence of 14.3% in aspirin users versus 8.5% in nonusers (RRUnivariate, 1.79; 95% CI, 1.04-3.09; \( P = .036 \) and RRMultivariable, 3.82; 95% CI, 1.95-7.5; \( P < .001 \)).

## DISCUSSION

This study shows that aspirin use after diagnosis is associated with longer survival. This association persisted after adjustment for age, sex, stage, treatment, diabetes mellitus, dyslipidemia, hypertension, and cardiovascular diseases at time of diagnosis. The potential benefit from aspirin was particularly observed in stages II and III. The results in stage IV were not statistically significant. Given the lower number of patients in stage IV, larger studies are warranted to determine the significance of aspirin in stage IV melanoma. In early stages, existing interventions that mostly involve surgery are largely curative, so mortality from melanoma is low regardless of aspirin use. However, in stages II and III, aspirin could potentially be a potent adjuvant therapy improving survival. In a recent meta-analysis, pooled data from randomized clinical trials (designed for cardiovascular end points) and observational studies showed that aspirin was associated with longer survival in colon, prostate, and breast cancers.18

Another interesting finding is that aspirin use before diagnosis corresponded to earlier stage of melanoma. This association is supported by existing evidence regarding the role of platelets in the metastasis process, including creating a niche for tumor cells in the target organs.19,20 We analyzed other tumor parameters such as Breslow thickness greater than 1 mm, mitotic rate, lymphovascular invasion, and primary tumor regression and saw no advantage with aspirin use. A smaller study, however, has demonstrated that prediagnosis aspirin use was associated with smaller Breslow thickness.15 Further studies are needed to resolve this discrepancy and exclude the potential of publication bias. On the other hand, ulceration was more common among aspirin users, which could be related to more bleeding diathesis with aspirin. Given the retrospective nature of this study, the correlation with an earlier stage should be interpreted with caution. It is

| Table III. Postdiagnosis aspirin use and overall survival rates (%) at different time points stratified by stage |
|--------------------------|--------------------------|--------------------------|--------------------------|
| Stage/time               | No aspirin | Aspirin |
| In situ 3 y              | 95         | 94        |
| 5 y                    | 92         | 89        |
| 7 y                    | 90         | 88        |
| Stage I 3 y              | 97         | 95        |
| 5 y                    | 94         | 88        |
| 7 y                    | 92         | 87        |
| Stage II 3 y             | 85         | 85        |
| 5 y                    | 76         | 82        |
| 7 y                    | 68         | 79        |
| Stage III 3 y            | 72         | 85        |
| 5 y                    | 61         | 77        |
| 7 y                    | 51         | 69        |
| Stage IV 3 y             | 14         | 23        |
| 5 y                    | 13         | 23        |
| 7 y                    | 13         | 23        |

**Fig 1.** Melanoma, aspirin use, and overall survival. Kaplan-Meier curves show longer overall survival in patients with stage II to IV melanoma who are using aspirin than in nonusers. Aspirin use is not associated with survival in stage 0 or I patients. Survival curves were compared by using log-rank tests. Stage 0 or I: unadjusted hazard ratio, 1.30; 95% confidence interval, 0.85-1.98; \( P = .23 \). Stage II to IV: unadjusted hazard ratio, 0.58; 95% confidence interval, 0.41-0.80; \( P = .001 \).
possible that patients with closer medical care are more likely to be prescribed aspirin. This closer follow up could in turn translate to earlier detection of melanoma. However, given the recognized role of platelets in metastasis, it is conceivable that targeting them with aspirin could retard cancer dissemination and result in diagnosis at an earlier stage.

Many of the patients with aspirin intake after diagnosis might have also taken aspirin before diagnosis. Arguably, the period of aspirin intake before diagnosis could modify the developing melanoma and potentially affect survival irrespective of postdiagnosis aspirin use. This point was addressed here through 2 approaches. First, we checked whether aspirin use before diagnosis correlated with good prognostic histologic features and stage. Prediagnosis aspirin was associated with earlier stage; however, stage was adjusted for in the multivariable regression models. In the second approach, patients who received aspirin after diagnosis were stratified into those who also took aspirin before diagnosis and those who did not. The risk reduction was comparable in the 2 strata.

The protective effect of aspirin in melanoma could be explained by cancer-intrinsic and cancer-extrinsic factors (Fig 2). We recently showed that platelets suppress T-cell immunity against melanoma and antiplatelet agents such as aspirin improved the efficacy of antimelanoma immune response. Aspirin also exerts direct antiproliferative effects on melanoma cells in vitro, partly by inducing oxidative stress, and cyclooxygenase-2 is overexpressed in cutaneous melanomas compared with benign nevi. In addition, platelets are involved in cancer progression and metastasis through the secretion of molecules such as transforming growth factor-β (TGFβ), vascular endothelial growth factor, and metalloproteinases. For example, platelet-derived TGFβ induces the TGFβ/Smad and nuclear factor κ-light chain enhancer of activated B cells pathways in cancer, inducing metastasis. Platelets also upregulate c-myc and cancer cell proliferation, a process inhibited by aspirin. In melanoma, platelets facilitate metastasis to the lungs; the mechanisms include fibrinogen-dependent protection from natural killer cells. Platelet-derived prostaglandin F2α (PGF2α) mediates the activation of acid sphingomyelinase, or C-type lectin-type receptor 2–mediated platelet activation. Thus, platelets could be a strategic target in the treatment of melanoma.

This retrospective study has some limitations. The optimal time frame and duration of aspirin use relative to conventional treatments could not be elucidated. In addition, the use of aspirin was confounded by multiple variables: it was associated with older age, cardiovascular comorbidities, and male sex. Given that older age and male sex are associated with shorter survival in melanoma and cardiovascular diseases decrease life expectancy and thus survival from time of diagnosis, univariate analysis likely diminishes the association between aspirin use and survival. Given the high correlation between aspirin use and cardiovascular comorbidities, controlling for them does not entirely resolve this bias. The retrospective and observational nature of the study also harbors intrinsic limitations related to data accuracy and inability to establish causality. Moreover, our study is limited to white patients, as this demographic accounts for the vast majority of melanoma cases. Previous studies have demonstrated better outcomes in whites than in other races. Given the stark differences in survival by stage, a stratified analysis is...
warranted. However, the small number of nonwhite patients prohibited such an analysis.

A clinical trial is currently ongoing to assess the impact of long-term aspirin intake on recurrence and survival in colorectal, gastrointestinal, prostate, and breast cancers, and a similar trial in melanoma is warranted. However, the small number of nonwhite patients receiving immunotherapy displayed a benefit from aspirin comparable to that of other patients in stages III and IV. Thus, future trials should evaluate the utility of combining aspirin with immunotherapy, a modality that is increasingly being implemented as first-line treatment in advanced melanoma.

We are indebted to the Simon Cancer Center registry and the Clinical Data Warehouse at Indiana University Health for providing the patient data used in this study.

REFERENCES


Supplemental Table I. Aspirin is associated with overall survival in patients of all races combined

<table>
<thead>
<tr>
<th>Variable</th>
<th>No aspirin</th>
<th>Aspirin</th>
<th>P value</th>
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</thead>
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<td>HR&lt;sub&gt;Unadjusted&lt;/sub&gt; (95% CI)</td>
<td>1.0</td>
<td>0.65 (0.51-0.85)</td>
<td>.001</td>
</tr>
<tr>
<td>HR&lt;sub&gt;Adjusted&lt;/sub&gt; * (95% CI)</td>
<td>1.0</td>
<td>0.57 (0.44-0.74)</td>
<td>.001</td>
</tr>
</tbody>
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CI, Confidence interval; HR, hazard ratio.
*HR adjusted for age, sex, stage, and treatment modality.