Aspirin use and ovarian cancer mortality in a Danish nationwide cohort study

Freija Verdoodt1, Susanne K Kjaer1,2, Christian Dehlendorff3 and Søren Friis*,3,4

1Unit of Virus, Lifestyle and Genes, Danish Cancer Society Research Center, Strandboulevarden 49, Copenhagen 2100, Denmark; 2Department of Gynaecology, Rigshospitalet, University of Copenhagen, Blegdamsvej 9, Copenhagen 2100, Denmark; 3Unit of Statistics and Pharmacoepidemiology, Danish Cancer Society Research Center, Strandboulevarden 49, Copenhagen 2100, Denmark and 4Department of Public Health, University of Copenhagen, Øster Farimagsgade 5, Copenhagen 1014, Denmark

Background: Increasing data suggest that aspirin use may improve cancer survival; however, the evidence is sparse for ovarian cancer.

Methods: We examined the association between postdiagnosis use of low-dose aspirin and mortality in a nationwide cohort of women with epithelial ovarian cancer between 2000 and 2012. Information on filled prescriptions of low-dose aspirin, dates and causes of death, and potential confounding factors was obtained from nationwide Danish registries. We used Cox regression models to estimate hazard ratios (HRs) with 95% confidence intervals (CIs) for ovarian cancer-specific or other-cause mortality associated with low-dose aspirin use.

Results: Among 4117 patients, postdiagnosis use of low-dose aspirin was associated with HRs of 1.02 (95% CI: 0.87–1.20) for ovarian cancer mortality and 1.06 (95% CI: 0.77–1.47) for other-cause mortality. Hazard ratios remained neutral according to patterns of low-dose aspirin use, including prediagnosis use or established mortality predictors.

Conclusions: Low-dose aspirin use did not reduce mortality among ovarian cancer patients.

Despite some advances in treatment modalities, the survival of ovarian cancer has hardly improved for decades and identification of modifiable factors that can improve the prognosis of ovarian cancer patients remains a high priority (Allemani et al, 2015).

Several epidemiologic studies suggest improved cancer outcomes with regular aspirin use among patients with clinically manifest cancer, and a number of randomised clinical trials are currently ongoing to evaluate the role of aspirin in the treatment of common cancers, notably colorectal cancer (Coyle et al, 2016; Elwood et al, 2010). The exact mechanisms behind the anti-neoplastic effects of aspirin remain to be established (Thun et al, 2012; Umar et al, 2016). For ovarian cancer, some studies in murine models and human cell lines have demonstrated an interaction between platelets and proliferation, angiogenesis, and metastasis of ovarian tumours, suggesting a role for aspirin via the antiplatelet effect (Cooke et al, 2015; Cho et al, 2017); however, other mechanisms have also been suggested (Hudson et al, 2008; Gates et al, 2010). Only few epidemiologic studies of ovarian cancer patients have evaluated outcomes associated with aspirin use and the results have been too equivocal to allow efficient design of clinical trials (Minlikeeva et al, 2015; Nagle et al, 2015; Bar et al, 2016; Dixon et al, 2017; Verdoodt et al, 2017b). A pooled analysis of 12 studies within the Ovarian Cancer Association Consortium (OCAC) reported a neutral association between aspirin use and overall survival among ovarian cancer patients; however, aspirin exposure was self-reported and only prediagnosis use was evaluated (Dixon et al, 2017). The influence of postdiagnosis aspirin use, a clinically more relevant exposure, has been explored in only one small cohort study reporting a statistically significant 50% reduction in overall mortality with aspirin use (Bar et al, 2016). This prompted us to conduct a cohort study of postdiagnosis low-dose aspirin use and mortality among ovarian cancer patients in Denmark, using the unique Danish nationwide registries.
MATERIALS AND METHODS

From the Danish Cancer Registry, we identified all women aged 30–84 years with incident primary epithelial ovarian cancer between 2000 and 2012 and no history of cancer (except non-melanoma skin cancer). Information on filled prescriptions for low-dose aspirin and other drugs, tumour and patient characteristics, comorbid conditions, and mortality outcomes were retrieved from nationwide demographic, prescription, and patient registries, using the unique civil registration number assigned to all Danish residents for linkage. The Supplementary Material provides a detailed description of the registries, with codes for ovarian cancer, drug exposure, and covariates.

The study outcomes were ovarian cancer-specific and other-cause mortality. Patients were followed from 1 year after ovarian cancer diagnosis until death, migration, or end of the study (31 December 2013).

We defined postdiagnosis use of low-dose (75–150 mg) aspirin as ≥1 prescription filled after the ovarian cancer diagnosis. Prediagnosis use of low-dose aspirin was defined as ≥1 prescription within 5 years before the ovarian cancer diagnosis. In the primary analysis, we assessed postdiagnosis use of low-dose aspirin as a time-varying covariate lagged by 1 year (Chubak et al., 2013). Thus, postdiagnosis low-dose aspirin users were regarded as non-users until 1 year after their first prescription.

In secondary analyses, we evaluated the influence of timing of low-dose aspirin use by developing a supplementary time-varying exposure matrix: (1) no pre- or postdiagnosis use (reference group), (2) prediagnosis use only, (3) pre- and postdiagnosis use, and (4) postdiagnosis use only.

In two sensitivity analyses with fixed exposure periods, low-dose aspirin use was assessed from time of diagnosis until the start of follow-up at 1 or 3 years following the ovarian cancer diagnosis, and was considered invariable thereafter (Verdoodt et al., 2017a).

We used Cox proportional hazard regression models to estimate hazard ratios (HRs) and 95% confidence intervals (CIs) for the association between postdiagnosis low-dose aspirin use and ovarian cancer-specific and other-cause mortality. Minimally adjusted analyses included age at diagnosis, clinical stage, and year of diagnosis. Fully adjusted models further included tumour histology, chemotherapy, highest achieved education, disposable income, marital status, comorbid conditions, and non-aspirin drug use (Table 1 and Supplementary Material). The proportional hazards assumption was tested using scaled Schoenfeld residuals. Finally, we evaluated the influence of competing risks as a result of death from other causes using the subdistribution hazards model proposed by Fine and Gray adapted for time-dependent covariates (Fine and Gray, 1999).

All analyses were performed using the R statistical software version 3.2.3 and the survival package (R Foundation for Statistical Computing, 2015; Therneau, 2015). STROBE guidelines were used to outline this study (von Elm et al., 2007). The Danish Data Protection Agency and Statistics Denmark’s Scientific Board approved the study. According to Danish law, ethical approval is not required for registry-based studies (Thygesen et al., 2011).

RESULTS

Among 5439 eligible women with a primary diagnosis of epithelial ovarian cancer, 4117 were alive 1 year after the diagnosis and included in our study. During a mean follow-up of 3.6 years (maximum 13 years), 2245 (55%) patients died and of these, 1903 (85%) women died from ovarian cancer. Characteristics of the study population are shown in Table 1.

In the primary, time-varying analysis, we saw no association between postdiagnosis use of low-dose aspirin and ovarian cancer (HR: 1.02, 95% CI: 0.87–1.20) or other-cause (HR: 1.06, 95% CI: 0.77–1.47) mortality (Table 2). Further, we observed no substantial variation in HRs according to estimated dose (tablet size), cumulative amount of postdiagnosis low-dose aspirin use, or with timing of use (Table 2). Stratification according to tumour histology (Table 3), age at diagnosis, clinical stage, or year of diagnosis (Online Supplementary eTable 2) did not materially influence the associations.

We also observed overall neutral associations for ovarian cancer-specific and other-cause mortality in sensitivity analyses, except for an increased HR with short duration of low-dose aspirin use in the 3-year analysis; however, these estimates were based on small numbers (Supplementary eTable 3). Finally, analyses accounting for competing risks (Fine and Gray) exhibited results similar to those of the primary analyses (data not shown).

DISCUSSION

Our finding of a null association between use of low-dose aspirin and mortality after ovarian cancer is compatible with the results of the OCAC study based on prediagnosis use, but our results are in contrast to a previous study reporting a substantial reduction in overall mortality among ovarian cancer patients with postdiagnosis aspirin use (Bar et al., 2016). However, besides a small sample size, the latter cohort study was prone to time-related bias which are likely to have influenced the estimates (Chubak et al., 2013).

In our study, we evaluated the influence of low-dose aspirin on mortality after ovarian cancer, assuming that one tablet was equivalent to daily use. Higher dosages of aspirin might be required to obtain a beneficial effect on ovarian cancer prognosis; however, this is not readily supported by analyses of various patterns of low-dose aspirin use in our study, or the similar associations for low-dose (<100 mg) and higher-dose (≥100 mg) aspirin in the OCAC study (Dixon et al., 2017). Moreover, for cancer in general, there is no solid evidence that doses of aspirin higher than those used in cardioprotection (75–150 mg) would provide stronger anticancer effects (Coyle et al., 2016; Elwood et al., 2016).

Among the strengths of our study were the nationwide cohort, large study size, high-quality and continuously updated registry data, and complete follow-up. The use of the Danish Prescription Registry ensured complete assessment of prescription drug use. The study design eliminated recall bias, and minimised selection bias and time-related biases.

A limitation of our study was the lack of data on over-the-counter (OTC) purchases of aspirin. However, in Denmark, most (>90%) of the total sales of low-dose aspirin are prescribed (Schmidt et al., 2014), and this proportion may even be higher in cancer patients who are typically under close medical surveillance. High-dose (500 mg) aspirin preparations are mainly sold OTC in Denmark, and thus use of aspirin at this dose may have driven a possible slight inverse association towards the null given that high-dose aspirin is more likely to be used by non-users of low-dose aspirin than among users. However, high-dose aspirin is mainly used for short-term treatment of transient, non-cancer-related pain and therefore OTC use of high-dose aspirin likely resulted in at most minor misclassification of long-term aspirin use. Moreover, the absence of any material differences in associations with increasing dose and cumulative amount of low-dose aspirin indicates that OTC sales of high-dose aspirin likely did not have major impact on our results. Furthermore, in Denmark, regular use of drugs, including high-dose aspirin, is generally prescribed...
because of at least 50% cost reimbursement and the need for medical surveillance for adverse effects (Schmidt et al., 2014). In our study population, only five patients filled a minimum of one prescription for high-dose aspirin, thus suggesting that use of aspirin at this dose was indeed only sporadic. Still, although we adjusted for several potential confounding factors, residual

| Table 1. Characteristics of ovarian cancer patients surviving at least 1 year after the ovarian cancer diagnosis, according to post-diagnosis use of low-dose aspirin within the first year after diagnosis |
|-----------------|-----------------|-----------------|
|                  | Non-users (n = 3650) | Post-diagnosis low-dose aspirin users (n = 467) |
|                  | No. (%)           | No. (%)         |
| Prediagnosis low-dose aspirin use |                  |                 |
| Use             | 179 (4.9)         | 374 (80.1)      |
| Non-use         | 3471 (95.1)       | 93 (19.9)       |
| Year of diagnosis |                  |                 |
| 2000–2003       | 1197 (32.8)       | 130 (27.8)      |
| 2004–2007       | 1131 (31.0)       | 123 (26.3)      |
| 2008–2012       | 1322 (36.2)       | 214 (45.8)      |
| Age at diagnosis |                  |                 |
| Median (IQR)    | 60 (52–68)        | 70 (63–76)      |
| Clinical stage  |                  |                 |
| Localised       | 1470 (40.3)       | 171 (36.6)      |
| Non-localised   | 1913 (52.4)       | 247 (52.9)      |
| Unknown         | 267 (7.3)         | 49 (10.5)       |
| Tumour histology |                  |                 |
| Serous          | 2164 (59.3)       | 279 (59.7)      |
| Endometrioid    | 484 (13.3)        | 72 (15.4)       |
| Mucinous        | 351 (9.6)         | 28 (6.0)        |
| Clear cell      | 204 (5.6)         | 20 (4.3)        |
| Other           | 447 (12.2)        | 68 (14.6)       |
| Chemotherapy    |                  |                 |
| Yes             | 2814 (77.1)       | 366 (78.4)      |
| No              | 836 (22.9)        | 101 (21.6)      |
| Highest achieved education |        |                 |
| Basic           | 99 (2.7)          | 11 (2.4)        |
| Vocational/short| 2560 (70.1)       | 388 (83.1)      |
| Long/medium     | 898 (24.6)        | 54 (11.6)       |
| Unknown         | 93 (2.5)          | 14 (3.0)        |
| Disposable income |              |                 |
| Low             | 1088 (29.8)       | 190 (40.7)      |
| Medium          | 1208 (33.1)       | 186 (39.8)      |
| High            | 1354 (37.1)       | 91 (19.5)       |
| Marital status  |                  |                 |
| Divorced        | 460 (12.6)        | 51 (10.9)       |
| Married         | 2274 (62.3)       | 242 (51.8)      |
| Unmarried       | 418 (11.5)        | 35 (7.5)        |
| Widow           | 498 (13.6)        | 139 (29.8)      |
| Comorbid conditions |            |                 |
| Diabetes mellitus | 137 (3.8)       | 69 (14.8)       |
| COPD            | 134 (3.7)         | 37 (7.9)        |
| Ischaemic heart disease | 113 (3.1) | 137 (29.3)    |
| Congestive heart disease | 45 (1.2) | 32 (6.9)       |
| Cerebrovascular disease | 107 (2.9) | 85 (18.2)      |
| Atrial fibrillation | 92 (2.5)         | 50 (10.7)       |
| Other drug use (≥1 post-diagnosis prescription) | | |
| Non-aspirin NSAIDs | 1050 (28.8) | 157 (33.6) |
| Antiplatelet drugs (other)* | 32 (0.9) | 55 (11.8) |
| Anticoagulants (other)b | 227 (6.2) | 38 (8.1) |
| Statins         | 287 (7.9)         | 186 (39.8)      |
| β-Blockers      | 300 (8.2)         | 166 (35.5)      |
| Calcium channel blockers | 301 (8.2) | 125 (26.8) |
| ACE inhibitors  | 249 (6.8)         | 115 (24.6)      |
| ARBs            | 221 (6.1)         | 67 (14.3)       |
| Antihypertensives (other)f | 915 (25.1) | 232 (49.7) |
| Cardiovascular drugs (other)d | 83 (2.3) | 79 (16.9) |
| Insulin and analogues | 38 (1.0) | 27 (5.8) |
| Metformin       | 56 (1.5)          | 31 (6.6)        |
| Oral antidiabetics (other)* | 52 (1.4) | 24 (5.1) |
| Paracetamol     | 852 (23.3)        | 195 (41.8)      |
| Proton pump inhibitors | 988 (27.1) | 192 (41.1) |
| Bisphosphonates | 101 (2.8)         | 26 (5.6)        |
| Antihistamines  | 268 (7.3)         | 41 (8.8)        |
| Drugs against COPD | 40 (1.1) | 15 (3.2) |
| High-dose aspirinf | 5 (0) | 0 (0) |

Abbreviations: ACE — angiotensin-converting enzyme; ADP — adenosine diphosphate; ARB — angiotensin II receptor blocker; COPD — chronic obstructive pulmonary disease; IQR — interquartile range; NSAIDs — non-steroidal anti-inflammatory drugs.

*Dipyridamole and ADP receptor antagonists.

bVitamin K antagonists, heparin group, direct thrombin inhibitors, and direct factor Xa inhibitors.

cAntidiabetic drugs and diuretics.

dCardiac glycosides, antiarrhythmic agents, cardiac stimulants, vasodilators, and prostaglandins.

*eSulphonylureas, α-glucosidase inhibitors, thiazolinediones, dipeptidyl-peptidase-4 inhibitors, and other blood glucose-lowering drugs.

fNot included in multivariable-adjusted analysis due to low numbers.
confounding could have been introduced by lifestyle factors, such as physical activity, smoking and obesity, or other unmeasured factors potentially associated with both low-dose aspirin use and ovarian cancer mortality.

In conclusion, we found no evidence of reduced mortality among ovarian cancer patients associated with postdiagnosis use of low-dose aspirin.

Table 2. Association between postdiagnosis low-dose aspirin use and ovarian cancer-specific and other-cause mortality, using time-varying analysis

| Low-dose aspirin | Ovarian cancer-specific mortality | Other-cause mortality |
|------------------|----------------------------------|----------------------|
|                  | Person years | Deaths | HR basic adjustmenta (95% CI) | HR full adjustmentb (95% CI) | Person years | Deaths | HR basic adjustmenta (95% CI) | HR full adjustmentb (95% CI) |
| Non-use          | 12914        | 1661   | 1 | 1 | 12914 | 272   | 1 | 1.24 (0.93-1.66) | 1.06 (0.77–1.47) |
| Post-diagnosis use | 1832        | 242    | 1.07 (0.93–1.23) | 1.02 (0.87–1.20) | 1832 | 70    | 1 | 1.24 (0.93-1.66) | 1.06 (0.77–1.47) |

Dose (tablet size)

| 75–100 mg | 1368 | 179 | 1.03 (0.88–1.21) | 0.98 (0.82–1.17) | 1368 | 45 | 1.08 (0.77–1.51) | 0.95 (0.66–1.38) |
| 150 mg | 325 | 52 | 1.23 (0.93–1.64) | 1.20 (0.90–1.61) | 325 | 14 | 1.52 (0.86–2.68) | 1.31 (0.73–2.37) |
| Mixed | 139 | 11 | 1.07 (0.87–1.37) | 0.97 (0.74–1.31) | 139 | 11 | 2.28 (1.16–4.48) | 1.53 (0.74–3.15) |

Cumulative amount (tablets)

| 1–365 | 620 | 114 | 1.12 (0.92–1.37) | 0.87 (0.67–1.11) | 620 | 25 | 1.45 (0.94-2.22) | 1.29 (0.83–2.00) |
| 366–1095 | 681 | 78 | 0.89 (0.71–1.13) | 0.76 (0.57–1.02) | 681 | 23 | 1.19 (0.76–1.84) | 0.79 (0.54–1.22) |
| >1096 | 531 | 50 | 1.34 (0.98–1.84) | 1.22 (0.88–1.70) | 531 | 22 | 1.07 (0.64–1.77) | 0.87 (0.50–1.49) |

Timing

| Never use | 12378 | 1552 | 1 | 1 | 12378 | 253 | 1 | 1 | 1.11 (0.68–1.80) | 0.91 (0.54–1.51) |
| Prediagnosis use only | 536 | 109 | 1.03 (0.84–1.26) | 0.92 (0.74–1.13) | 536 | 19 | 1.11 (0.68–1.80) | 0.91 (0.54–1.51) |
| Post-diagnosis use only | 841 | 74 | 1.07 (0.83–1.36) | 1.00 (0.77–1.29) | 841 | 26 | 0.99 (0.63–1.56) | 0.89 (0.56–1.42) |
| Pre- and postdiagnosis use | 992 | 168 | 1.07 (0.91–1.27) | 1.01 (0.84–1.22) | 992 | 44 | 1.44 (1.02–2.03) | 1.18 (0.79–1.75) |

Prediagnosis cumulative amount (tablets)

| 1–999 | 506 | 80 | 0.99 (0.79–1.25) | 0.95 (0.74–1.22) | 506 | 17 | 1.15 (0.69–1.91) | 1.01 (0.58–1.75) |
| >1000 | 486 | 88 | 1.16 (0.93–1.44) | 1.07 (0.84–1.36) | 486 | 27 | 1.72 (1.12–2.63) | 1.34 (0.82–2.17) |

Abbreviations: CI = confidence interval; HR = hazard ratio.
*aAdjusted for age at diagnosis, year of diagnosis, and clinical stage.
*bAdjusted for age at diagnosis, year of diagnosis, clinical stage, tumour histology, chemotherapy, highest achieved education, disposable income, marital status, use of non-aspirin drugs, and comorbid conditions.

cA supplementary exposure matrix including both pre- and postdiagnosis low-dose aspirin use was developed, using four time-varying categories: (1) no pre- or postdiagnosis use (‘never use’, reference), (2) prediagnosis use only, (3) postdiagnosis use only, and (4) both pre- and postdiagnosis use.

dEvaluation according to cumulative number of prediagnosis low-dose aspirin tablets among patients with both pre- and postdiagnosis use, compared with never use.

Table 3. Association between post-diagnosis low-dose aspirin use and ovarian cancer-specific and other-cause mortality, using time-varying analysis and stratified by tumour histology

| Tumour histology | Ovarian cancer-specific mortality | Other-cause mortality |
|------------------|----------------------------------|----------------------|
|                   | Person years | Deaths | HR basic adjustmenta (95% CI) | HR full adjustmentb (95% CI) | Person years | Deaths | HR basic adjustmenta (95% CI) | HR full adjustmentb (95% CI) |
| Serous Non-use | 6643 | 917 | 1123 | 1.04 (0.87–1.24) | 0.98 (0.81–1.19) | 6643 | 917 | 150 | 1.22 (0.80–1.86) | 0.95 (0.61–1.50) |
| Post-diagnosis use | 2209 | 332 | 153 | 1.30 (0.82–2.07) | 1.26 (0.79–2.02) | 2209 | 332 | 35 | 1 | 0.49 (0.19–1.24) | 0.50 (0.20–1.30) |
| Endometrioid Non-use | 1757 | 238 | 64 | 1.06 (0.44–2.56) | 0.92 (0.37–2.25) | 1757 | 238 | 34 | 1.91 (0.85–4.30) | 1.52 (0.66–3.48) |
| Post-diagnosis use | 803 | 151 | 71 | 0.68 (0.30–1.55) | 0.72 (0.31–1.63) | 803 | 151 | 11 | 2.46 (0.57–10.60) | 1.87 (0.45–7.86) |
| Mucinous Non-use | 1502 | 194 | 250 | 1.06 (0.70–1.59) | 1.02 (0.67–1.54) | 1502 | 194 | 42 | 1 | 1.43 (0.60–3.42) | 1.37 (0.55–3.42) |

Abbreviations: CI = confidence interval; HR = hazard ratio.
*aAdjusted for age at diagnosis, year of diagnosis, and clinical stage.
*bAdjusted for age at diagnosis, year of diagnosis, clinical stage, chemotherapy, highest achieved education, disposable income, marital status, use of non-aspirin drugs, and comorbid conditions.

Acknowledgements

This project was funded by the Sapere Aude-program of the Danish Council for Independent Research (Det Frie Forskningsråd Sapere Aude-program, project No. 6110-00596B), and the Mermaid project (Mermaid III).
CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

Allemanni C, Weir HK, Carreira H, Harewood R, Spika D, Wang XS, Bannon F, Ahn JV, Johnson CJ, Bonaventure A, Marcos-Gragera R, Stiller C, Azevedo e Silva G, Chen WQ, Ogungbiyi OJ, Rachel B, Soeberg MJ, You H, Matsuda T, Bielska-Lasota M, Storm H, Tucker TC, Coleman MP (2015) Global surveillance of cancer survival 1995–2009: analysis of individual data for 25,676,887 patients from 279 population-based registries in 67 countries (CONCORD-2). Lancet 385: 977–1010.

Bar D, Lavei O, Stein N, Feferkorn I, Shai A (2016) The effect of metabolic comorbidities and commonly used drugs on the prognosis of patients with ovarian cancer. Eur J Obstet Gynecol Reprod Biol 207: 227–231.

Cho MS, Noh K, Haemmerle M, Li D, Park H, Hu Q, Hisamatsu T, Allemani C, Weir HK, Carreira H, Harewood R, Spika D, Wang XS, Tucker TC, Coleman MP (2015) Global surveillance of cancer survival 1995–2009: analysis of individual data for 25,676,887 patients from 279 population-based registries in 67 countries (CONCORD-2). Lancet 385: 977–1010.

This work is published under the standard license to publish agreement. After 12 months the work will become freely available and the license terms will switch to a Creative Commons Attribution-NonCommercial-Share Alike 4.0 Unported License.

Supplementary Information accompanies this paper on British Journal of Cancer website (http://www.nature.com/bjc)