Use of disulfiram and risk of cancer: a population-based case–control study

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Experimental studies have indicated that disulfiram (Antabuse) has antineoplastic effects against melanoma, breast, and prostate cancer. To explore this hypothesis, we examined the association between disulfiram use and these cancers in a nationwide register-based case–control study nested within ever-users (≥ one prescription) of disulfiram. Cases were all Danish individuals with a histologically verified first-time diagnosis of malignant melanoma, breast, or prostate cancer during 2000–2009. For each case, we selected four cancer-free controls matched for age, sex, and year of first disulfiram prescription using risk set sampling. Similarly, for secondary analyses, we selected case–control populations for selected tobacco-related and alcohol-related cancer types, that is, cancers of the buccal cavity, liver, lung, and colorectal cancer. Disulfiram use 1 year before cancer diagnosis and the corresponding date for controls were disregarded. We estimated crude and adjusted odds ratios and 95% confidence intervals (CI) for cancer associated with long-term (≥ 500 daily defined doses) versus one-time (one prescription) use of disulfiram. Among 53 856 disulfiram users, we identified 166, 644, and 464 cases, respectively, of melanoma, breast, or prostate cancer. Adjusted odds ratios for melanoma, breast, or prostate cancer associated with long-term disulfiram use were 1.04 (95% CI: 0.60–1.78), 0.92 (95% CI: 0.70–1.22), and 0.77 (95% CI: 0.56–1.06), respectively. For prostate cancer, dose–response analyses showed a further risk reduction with the highest cumulative dose level of disulfiram; however, the test for trend did not reach statistical significance. Our study provides some epidemiological support for a protective effect of disulfiram against prostate and breast cancer. ***European Journal of Cancer Prevention*** **00:000–000 © 2013 Wolters Kluwer Health | Lippincott Williams & Wilkins.**

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Introduction

For more than 50 years, disulfiram (Antabuse) has been used in the treatment of alcohol dependence and is considered a well-tolerated drug with mild adverse effects (Borup *et al.*, 1992; Suh *et al.*, 2006). Disulfiram inhibits the enzyme aldehyde dehydrogenase, which induces increased acetaldehyde concentrations and unpleasant symptoms when alcohol is consumed. Laboratory studies have suggested that disulfiram may also inhibit the growth of in-situ breast cancer cells by blocking aldehyde dehydrogenase (Yip *et al.*, 2011). Other mechanisms have also been suggested. In particular, disulfiram has been found to inhibit the proteasome in melanoma, breast, and prostate cancer cell lines (Cen *et al.*, 2004; Chen *et al.*, 2005, 2006; Zhang *et al.*, 2010). Proteasome inhibition may promote antineoplastic activity by suppressing the nuclear factor-κB pathway because nuclear factor-κB activation is involved in carcinogenesis through expression of cell cycle genes, apoptosis inhibitors, and invasive proteases (Karin, 2006; Morrison *et al.*, 2010; Cvek, 2011). Moreover, disulfiram might inhibit DNA methylation in prostate cancer cells, which is considered an important step in the development of prostate cancer (Lin *et al.*, 2011).

To date, only a few human studies have investigated the potential antineoplastic effects of disulfiram, and the results have been conflicting (Verma *et al.*, 1990; Dufour *et al.*, 1993). As disulfiram is a safe and inexpensive drug, there is a strong interest in clarifying whether disulfiram has significant antineoplastic activity, and several on-going clinical trials are evaluating whether disulfiram can improve the prognosis in recurrent prostate cancer, metastatic melanoma, lung cancer, or in patients with liver metastasis (Lin and Lin, 2011). As disulfiram may inhibit cancer growth, we speculate that disulfiram could postpone the time to diagnosis or recurrence of cancer and in some individuals even avoid cancer diagnosis or death during lifetime. However, so far, no observational epidemiological study has evaluated whether disulfiram has cancer preventive properties.

To explore the associations between the long-term use of disulfiram and the risks of melanoma, breast, or

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prostate cancer in a large population-based setting, we carried out a nationwide case–control study in Denmark.

Materials and methods

Our study was designed as a case–control analysis nested within ever-users of disulfiram. The rationale behind this approach was that the use of disulfiram is strongly related to high consumption of alcohol, that is, the indication, as well as to tobacco smoking. As many cancers are alcohol or tobacco related, it is difficult to disentangle the intrinsic effect of disulfiram. Nesting our study within ever-users of disulfiram and having small-volume use as a reference addressed these problems.

Data sources

The Danish Cancer Registry has recorded incident cases of cancer in Denmark since 1943 and has been shown to have accurate and almost complete ascertainment of cancer cases (Storm *et al.*, 1997; Gjerstorff, 2011). Cancer diagnoses are recorded according to the International Classification of Diseases, version 10 (ICD-10), and the ICD for Oncology (ICD-O-1-3) for topography and morphology codes (Gjerstorff, 2011).

The Danish National Patient Register contains information on all nonpsychiatric hospital admissions in Denmark since 1977 and outpatient contacts since 1995. Hospital discharge diagnoses and ambulatory contact diagnoses have been coded according to ICD-8 from 1977 to 1993 and ICD-10 since 1994 (Lynge *et al.*, 2011).

The Danish National Prescription Registry contains data on all dispensed prescriptions since 1995 (Kildemoes *et al.*, 2011). Prescription data include the type of drug, date of dispensing, and quantity. Drugs are categorized according to the Anatomical Therapeutic Chemical (ATC) index and the quantity dispensed for each prescription is expressed by the defined daily dose (DDD) measure (WHO Collaborating Centre for Drug Statistics Methodology, 2012).

The Danish Civil Registration System has continuously updated data on the date of death and migration to and from Denmark (Pedersen, 2011).

The above registries were linked by the personal identification number, a unique identifier assigned to all Danish residents since 1968 that encodes sex and date of birth (Pedersen, 2011).

Finally, we obtained information from the 2010 Danish National Cohort Study (DANCOS), which contains detailed information on health characteristics, for example, lifestyle factors, medication use, and anthropometric measures, collected as part of the National Health Interview Survey (Davidsen *et al.*, 2011).

Study population

Eligible cases and controls were Danish residents who had filled at least one prescription of disulfiram 1 year before the index date, defined as the date of cancer diagnosis or the corresponding date for controls. Cases were individuals with a histologically verified diagnosis of malignant melanoma (ICD-10: C43), breast (C50), or prostate cancer (C61) during 2000–2009. We restricted the case population to individuals who had lived in Denmark continuously since 1995 and who had no history of cancer (except nonmelanoma skin cancer) before the index date.

Controls were selected using a risk set sampling strategy (Rothman *et al.*, 2008). For each case, we selected four controls fulfilling the above inclusion criteria for cases and matched by sex, birth year, and year of first disulfiram prescription. Patients were eligible for sampling as controls before they became cases. Thereby, the calculated odds ratios (ORs) were unbiased estimates of the incidence rate ratios that would have emerged from a cohort study in the same source population.

Exposure definition

One-time use of disulfiram (ATC: N07BB01) was defined as filling of a single prescription of less than or equal to 100 DDD disulfiram more than 1 year before the index date. Long-term use of disulfiram was defined as filling of at least 500 DDD more than 1 year before the index date. The 1-year lag-time was introduced to minimize possible selection bias (reverse causation) as drug use is known to increase within the last year before a cancer diagnosis (Jorgensen *et al.*, 2012).

Main analysis

The analysis was carried out as a conventional matched case–control study among users of disulfiram. ORs for the study cancers associated with the long-term use of disulfiram were calculated using conditional logistic regression to adjust for potential confounders. The main analysis was divided into two parts. First, long-term use of disulfiram was compared with one-time (i.e. one prescription) use. Second, duration–response associations were assessed by categorizing disulfiram use according to the cumulative amount dispensed more than 1 year before the index date, again as compared with one-time use.

As the potential confounders vary between cancer sites, we used a two-step approach, defining general and site-specific confounders. The prevalence (ever/never) of the following conditions was included in all regression models: chronic obstructive pulmonary disease as a crude marker of heavy smoking; any registered condition related to heavy alcohol abuse; and diabetes (composite measure of diabetes diagnoses or prescription of any antidiabetics). In addition, we included the Charlson comorbidity index (CCI) score and defined the level of comorbidity as none (CCI score: 0), low (CCI score: 1), or medium/high (CCI score: ≥ 2) (Charlson *et al.*, 1987; Thygesen *et al.*, 2011). Finally, we included highest completed education, categorized as (a) elementary school; (b) high school,

vocational education or short training; (c) medium-long training; or (d) missing or unknown (Jensen and Rasmussen, 2011).

Site-specific confounders were defined as the use of 5-α-reductase inhibitors in analyses of prostate cancer, hormone supplements in analyses of breast cancer and melanoma, and thiazides in melanoma analyses (Jensen *et al.*, 2008; Gupta and Driscoll, 2010; Chen, 2011; Azzouni and Mohler, 2012). All analyses were carried out using Stata Release 12.0 (StataCorp, College Station, Texas, USA).

Unmeasured confounding

Although we expected the nested analysis within ever-users of disulfiram to considerably reduce confounding by lifestyle factors, primarily the use of alcohol and tobacco, the possibility of residual confounding remained in comparisons of long-term versus one-time use of disulfiram. The direction of residual confounding by tobacco smoking and alcohol abuse in relation to adherence and duration of disulfiram use is difficult to predict. Frequent use of disulfiram might be associated with a high degree of abuse and therefore heavier drinking or smoking. The inverse might also be true, that is, adherence to disulfiram (i.e. alcohol abstinence) might be associated with less drinking or smoking as compared with one-time use. We used two different approaches to assess the direction and magnitude of this potential bias. First, we examined four cancer types associated with alcohol or tobacco use but with no acknowledged relation to use of disulfiram: (a) lung cancer (ICD-10: C33, C34, and C39) that is strongly related to smoking; (b) liver cancer (ICD-10: C22), related to alcohol and tobacco; (c) cancers of the buccal cavity or the pharynx (ICD-10: C00-C14 and C462), associated with both alcohol and tobacco, and (d) colorectal cancer (ICD-10: C18-20), associated with alcohol and weakly associated with tobacco (Baan *et al.*, 2007; Secretan *et al.*, 2009). For each of these cancer types, we repeated our duration–response analyses to assess whether long-term use of disulfiram influenced the risk of cancer.

Second, to directly assess the distribution of confounders between long-term and one-time users of disulfiram, we obtained data from the DANCOS survey conducted in 2010 (Davidsen *et al.*, 2011). Within this survey, we identified all respondents who had filled at least one prescription of disulfiram more than 1 year before the survey. To minimize prevalence bias, we excluded users who had filled a prescription less than 6 months before the survey. We then divided these individuals into one-time, intermediate, or long-term users of disulfiram using the same definitions as in the main analyses, and assessed the distributions of tobacco use, alcohol intake, and BMI within the three groups.

Sensitivity analyses

We also carried out a number of subgroup and sensitivity analyses using the same reference as in the main analyses (i.e. 1–100 DDD):

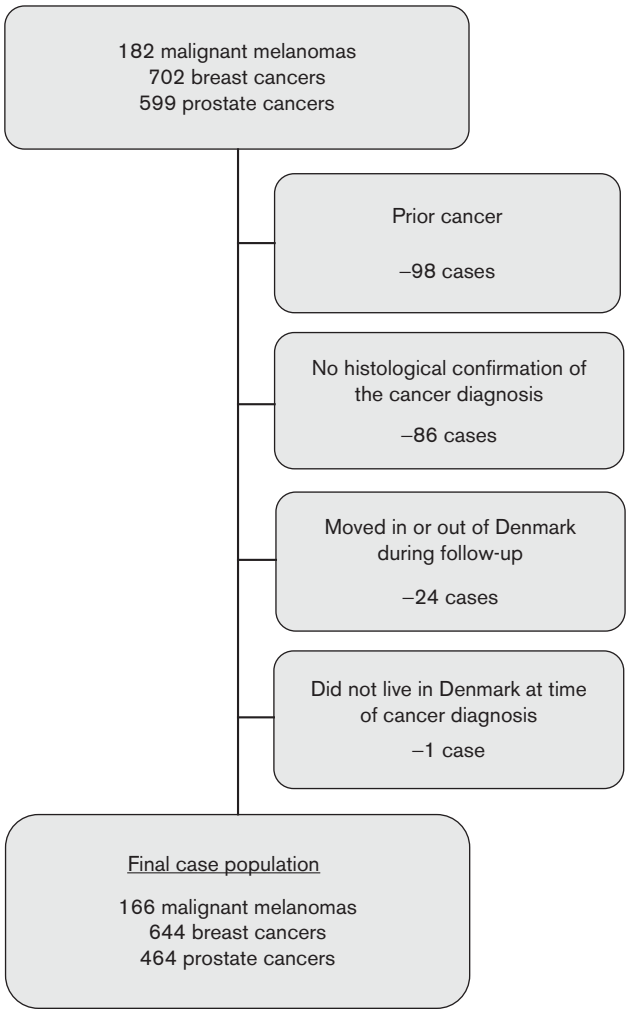
(1) We repeated the analyses stratified by sex and age (< 60 vs. ≥ 60 years).
(2) We repeated the analyses excluding (a) diabetic patients, (b) study participants using other drugs for alcohol dependence, and (c) study participants with any registered condition related to heavy alcohol abuse.
(3) As disulfiram has been marketed in Denmark since 1959, some misclassification of cumulative exposure was likely because of disulfiram use before the start of the Prescription Registry in 1995. We therefore applied a new-user design by excluding all cases (and all corresponding controls) and controls who had filled one or more prescription(s) of disulfiram within the period 1995–1996 (run-in period). To further evaluate the magnitude of misclassification because of left truncation, we carried out an exploratory analysis using the Odense Pharmacoepidemiological Database (OPED), a regional Danish prescription database covering a subregion of Denmark from 1990 (Gaist *et al.*, 1997). Within OPED, we identified a cohort of individuals with at least one prescription of disulfiram after 1995 and examined previous use of disulfiram during the period 1990–1994.
(4) We repeated the analyses after restricting disulfiram use to the period within 1–5 years before the index date.
(5) We changed the lag-time to 0–2 years.

Results

Among 53 856 eligible disulfiram users during 2000–2009, we identified 166, 644, and 464 cases of malignant melanoma, breast, or prostate cancer (Fig. 1). For three cases, no controls were eligible. The final study population thus constituted 1271 cases and 5046 controls (average 3.97 controls/case). The characteristics of cases and controls are shown in Table 1. No major differences were found between cases and controls in level of comorbidity, education, diagnoses, or drug use.

Among the 166 melanoma cases, 39 (23%) were long-term users of disulfiram, whereas 79 (48%) had filled only one prescription. The corresponding numbers among controls were 149 and 280, yielding a crude OR of 0.99 (95% confidence interval: 0.60–1.66) and an adjusted OR of 1.04 (0.60–1.78) (Table 2). For breast and prostate cancer, the adjusted ORs were 0.92 (0.70–1.22) and 0.77 (0.56–1.06), respectively. In dose–response analyses, the lowest ORs were found for prostate cancer associated with the highest cumulative dose of disulfiram, whereas no dose–response relationship was observed for

Fig. 1



Flow chart of case selection.

melanoma or breast cancer (Table 2). Test for trends showed no statistically significant associations.

The analysis of cancer sites associated with alcohol or tobacco use showed adjusted risk estimates close to unity for liver, lung, and colorectal cancer (Table 3). A decreased OR was observed for the buccal cavity/pharynx (OR 0.75; 0.58–0.99). Except for liver cancer, for which numbers were small, the lowest risk estimates were found with the highest cumulative exposure to disulfiram, although tests for trend only reached statistical significance for cancer of the buccal cavity and pharynx.

Table 4 presents the distribution of smoking habits, alcohol intake, and BMI in the DANCOS survey according to level of disulfiram exposure. No major differences were observed in BMI or smoking habits according to level of disulfiram use, whereas one-time users of disulfiram had markedly higher alcohol intake than those with intermediate or long-term use of disulfiram, who had larger proportions of alcohol abstainers.

Analyses stratified according to patient subgroups are shown in Table 5. No major effect measure modification was found with respect to sex, age, presence of diabetes, presence of conditions related to heavy alcohol intake, or use of other drugs to treat alcohol dependence.

The evaluation of potential misclassification of disulfiram use because of left truncation of drug exposure, using OPED data, showed that among individuals who had filled only one disulfiram prescription after January 1995, 17% had filled one or more prescriptions during 1990–1994. The corresponding prevalence was 38% among individuals who filled more than one prescription after January 1995.

Finally, sensitivity analyses of different lag periods (0–2 years), application of new-user design, or restriction of the exposure period to 1–5 years before the index date yielded results similar to those of the main analyses (data not shown).

Discussion

Our study indicates that disulfiram may have preventive effects against breast and prostate cancers, whereas our results did not point to an antineoplastic effect of disulfiram against malignant melanoma.

The main strengths of our study were the use of nationwide data resources covering the entire Danish population and a lengthy exposure period of up to 15 years. The validity of both exposure (disulfiram prescriptions) and outcome (cancer diagnoses) information was high, and also the covariates, that is, comorbidity, education, and concomitant drug use, were based on high-quality data (Gjerstorff, 2011; Jensen and Rasmussen, 2011; Kildemoes *et al.*, 2011; Lynge *et al.*, 2011; Thygesen *et al.*, 2011).

Our study also had some limitations. Misclassification because of in-hospital treatment (accounting for $\approx$15%) was most likely independent of case status, thus attenuating the risk estimates if present (Danish Health and Medicines Authority, 2011). Also, noncompliance with disulfiram was likely because of the relapsing and remitting nature of alcohol addiction. Poor compliance might have led to an overestimation of the cumulative disulfiram intake, also creating a potential conservative bias. Moreover, as disulfiram use before 1995 was quite frequent according to OPED data, it is conceivable that we somewhat underestimated disulfiram use among our study participants. Again, this potential misclassification was most likely nondifferential, that is, introducing a conservative bias.

The lack of information on lifestyle habits was also a potentially important limitation. However, for the cancer sites of primary interest, only breast cancer has an

Table 1 Characteristics of cancer cases and matched controls

	Cases ($n=1271$) [n (%)]	Controls ($n=5046$) [n (%)]
Men	588 (46.3)	2335 (46.3)
Women	683 (53.7)	2711 (53.7)
Age (median) (IQR, years)	61 (54–67)	60 (54–67)
Follow-up (median) (IQR, years)	6.9 (4.4–9.7)	7.0 (4.4–9.6)
Cancer site		
Melanoma	166 (13.1)	NA
Breast	642 (50.5)	NA
Prostate gland	463 (36.4)	NA
Use of disulfiram ≥ 1 year before the index date		
One-time use (1–100 DDD)	543 (42.7)	2031 (40.2)
Intermediate use (101–499 DDD)	425 (33.4)	1798 (35.6)
Long-term use (≥ 500 DDD)	303 (23.8)	1217 (24.1)
Charlson comorbidity index		
Score = 0	769 (60.5)	3062 (60.7)
Score = 1	300 (23.6)	1154 (22.9)
Score ≥ 2	202 (15.9)	830 (16.4)
Highest completed education		
Elementary school	476 (37.5)	2126 (42.1)
High school or short training	423 (33.3)	1654 (32.8)
Medium/long training	304 (23.9)	1008 (20.0)
Missing or unknown	68 (5.4)	258 (5.1)
Diagnoses		
Conditions related to alcohol	365 (28.7)	1471 (29.2)
Diabetes	93 (7.3)	434 (8.6)
COPD	95 (7.5)	424 (8.4)
Drugs[a]		
5-α-reductase inhibitors	1 (0.1)	13 (0.3)
Hormone replacement	232 (18.3)	808 (16.0)
Thiazides	168 (13.2)	691 (13.7)

COPD, chronic obstructive pulmonary disease; DDD, defined daily dose; IQR, interquartile range.
[a]Exposure defined by a cumulative use of ≥ 500 DDD before the index date.

Table 2 Association between disulfiram use and the risk of cancer, specified by cumulative use and cancer site

Cancer type	Cases exposed/unexposed	Controls exposed/unexposed	Crude OR	Adjusted OR[a]
Use ≥ 500 DDD				
Melanoma	39/79	149/280	0.99 (0.60–1.66)	1.04 (0.60–1.78)
Breast	159/269	603/1038	0.93 (0.71–1.22)	0.92 (0.70–1.22)
Prostate	105/195	465/713	0.77 (0.56–1.05)	0.77 (0.56–1.06)
Cumulative dose–response analysis				
Melanoma				
101–499 DDD	48/79	234/280	0.71 (0.46–1.08)	0.72 (0.46–1.13)
500–999 DDD	32/79	100/280	1.25 (0.71–2.21)	1.29 (0.71–2.36)
≥ 1000 DDD	7/79	49/280	0.59 (0.21–1.60)	0.64 (0.20–2.04)
Breast				
101–499 DDD	214/269	906/1038	0.93 (0.75–1.14)	0.91 (0.74–1.13)
500–999 DDD	106/269	384/1038	1.02 (0.75–1.40)	1.02 (0.74–1.41)
≥ 1000 DDD	53/269	219/1038	0.80 (0.50–1.29)	0.73 (0.44–1.20)
Prostate				
101–499 DDD	163/195	658/713	0.91 (0.71–1.16)	0.90 (0.70–1.16)
500–999 DDD	67/195	295/713	0.74 (0.51–1.07)	0.76 (0.52–1.10)
≥ 1000 DDD	38/195	170/713	0.84 (0.51–1.38)	0.86 (0.52–1.45)

COPD, chronic obstructive pulmonary disease, DDD, defined daily dose; OR, odds ratio.
[a]Adjusted for COPD; any registered condition related to heavy alcohol abuse; diabetes, Charlson comorbidity index and highest completed education. Furthermore, adjusted for use of 5-α-reductase inhibitors in analyses for prostate cancer, hormone supplements in analyses for breast cancer, and melanoma and thiazides in analyses for melanoma.

established relationship with alcohol consumption (Baan *et al.*, 2007). Moreover, tobacco smoking has not been associated convincingly with malignant melanoma, breast, or prostate cancer, and we do not suspect a large difference in exposure to UV-radiation according to disulfiram use (Secretan *et al.*, 2009; Leitzmann and Rohrmann 2012; Volkovova *et al.*, 2012).

The analyses applied to assess the residual confounding by alcohol and tobacco indicated that long-term users of disulfiram had lower alcohol consumption than study participants with only one prescription, whereas tobacco use was independent of the usage pattern of disulfiram. Long-term users of disulfiram had a lower incidence of cancer types known to be associated with alcohol, that is, cancers of the buccal cavity/pharynx and colorectum, compared with one prescription users. Also, in the DANCOS survey, long-term users of disulfiram showed lower alcohol consumption than single-prescription users, whereas tobacco use was independent of disulfiram use

Table 3 Sensitivity analyses, examining cancer sites associated with alcohol or tobacco consumption

Cancer type	Cases exposed/unexposed	Controls exposed/unexposed	Crude OR	Adjusted OR[a]
Use ≥ 500 DDD				
Buccal cavity and pharynx	166/335	745/1333	0.90 (0.70–1.15)	0.75 (0.58–0.99)
Liver	21/60	92/249	1.02 (0.53–1.95)	1.00 (0.48–2.10)
Lung	318/530	1286/2090	1.01 (0.84–1.22)	0.97 (0.80–1.17)
Colorectal	145/258	585/941	0.92 (0.70–1.21)	0.92 (0.70–1.22)
Duration–response analysis				
Buccal cavity and pharynx				
101–499 DDD	307/335	1145/1333	1.07 (0.88–1.28)	0.96 (0.79–1.17)
500–999 DDD	116/335	471/1333	0.98 (0.74–1.31)	0.82 (0.60–1.11)
≥ 1000 DDD	50/335	274/1333	0.71 (0.46–1.09)	0.54 (0.34–0.87)
Liver				
101–499 DDD	53/60	194/249	1.11 (0.71–1.74)	0.94 (0.57–1.54)
500–999 DDD	14/60	72/249	0.95 (0.46–1.98)	1.02 (0.43–2.39)
≥ 1000 DDD	7/60	20/249	1.44 (0.40–5.19)	1.20 (0.27–5.41)
Lung				
101–499 DDD	491/530	1958/2090	0.97 (0.84–1.12)	0.94 (0.81–1.10)
500–999 DDD	200/530	794/2090	1.03 (0.83–1.28)	0.99 (0.79–1.24)
≥ 1000 DDD	118/530	492/2090	0.93 (0.69–1.24)	0.90 (0.67–1.23)
Colorectal				
101–499 DDD	207/258	895/941	0.83 (0.67–1.03)	0.81 (0.65–1.01)
500–999 DDD	102/258	373/941	1.08 (0.79–1.47)	1.06 (0.77–1.46)
≥ 1000 DDD	43/258	212/941	0.65 (0.40–1.06)	0.62 (0.37–1.03)

COPD, chronic obstructive pulmonary disease; DDD, defined daily dose; OR, odds ratio.
[a]Adjusted for COPD; any registered condition related to heavy alcohol abuse; diabetes, Charlson comorbidity index and highest completed education.

Table 4 Distribution of potential confounders estimated from the Danish National Cohort Study survey 2010, specified by the amount of disulfiram filled >1 year before the index date

	n (%)			
	One-time users (*n* = 1475)	Intermediate users (*n* = 856)	Long-term users (*n* = 157)	OR[a]
Smoking status				
Never smoker	196 (13.3)	108 (12.6)	26 (16.6)	1.25
Exsmoker	350 (23.7)	215 (25.1)	33 (21.0)	0.89
Occasional smoker	48 (3.3)	20 (2.3)	2 (1.3)	0.39
Routine smoker	849 (57.6)	503 (58.8)	90 (57.3)	1.00 (reference)
Weekly alcohol consumption				
Not consumed for the last 12 months (U)	208 (14.1)	233 (27.2)	52 (33.1)	3.91
0	97 (6.6)	71 (8.3)	24 (15.3)	3.87
1–7	261 (17.7)	142 (16.6)	20 (12.7)	1.20
8–21	375 (25.4)	145 (16.9)	19 (12.1)	0.79
≥ 22	438 (29.7)	220 (25.7)	28 (17.8)	1.00 (reference)
BMI				
< 18.5	39 (2.6)	28 (3.3)	6 (3.8)	1.42
18.5–24.9	601 (40.8)	362 (42.3)	65 (41.4)	1.00 (reference)
25.0–29.9	532 (36.1)	280 (32.7)	55 (35.0)	0.96
≥ 30.0	258 (17.5)	153 (17.9)	25 (15.9)	0.90

One-time use, intermediate use, and long-term use were defined as a cumulative amount of ≤ 100, 101–499, and ≥ 500 DDD, respectively.
DDD, defined daily dose; OR, odds ratio.
[a]OR for the associations between long-term disulfiram use and the alcohol, smoking, or BMI indicators. One-time users constituted the reference group in all analyses.

(Davidsen *et al.*, 2011). Therefore, the decreased risk of breast cancer associated with the higher cumulative level of disulfiram might be confounded by lower alcohol consumption among long-term disulfiram users compared with single-prescription users. However, as alcohol consumption is not an established risk factor for prostate cancer, such confounding cannot explain the dose–response effect observed for prostate cancer (Leitzmann and Rohrmann, 2012).

According to the recommendations for the treatment of alcohol addiction, disulfiram should be administered in a supervised manner, that is, the patient should consume disulfiram at least twice a week in front of a health professional. As a result, the compliant disulfiram user will see a doctor on a regular basis, which may lead to surveillance bias. Surveillance bias might particularly affect long-term users of disulfiram and thus potentially lead to an underestimation of an antineoplastic effect of disulfiram. In Denmark, the incidence of prostate and breast cancer has increased markedly in the last 15 years because of, respectively, increased use of prostate-specific antigen tests and breast cancer screening programs (Jorgensen *et al.*, 2009; Outzen *et al.*, 2012). We thus conclude that the majority of biases that might relate to our study are conservative, that is, would tend to obscure or reduce a potential preventive effect. Finally, lack of statistical power limited our ability to draw firm conclusions.

Table 5 Associations between long-term use of disulfiram and the risk of cancer, specified by patient subgroups

Subgroups	Cases exposed/unexposed	Controls exposed/unexposed	Crude OR	Adjusted OR[a]
Male				
Melanoma	31/55	104/205	1.21 (0.67–2.18)	1.18 (0.63–2.20)
Prostate	105/195	465/713	0.77 (0.56–1.05)	0.77 (0.56–1.06)
Female				
Melanoma	8/24	45/75	0.53 (0.18–1.54)	0.67 (0.20–2.27)
Breast	158/265	596/1029	0.95 (0.72–1.25)	0.94 (0.71–1.25)
Age < 60 years				
Melanoma	22/48	92/184	0.94 (0.47–1.91)	0.95 (0.45–2.00)
Breast	78/165	293/658	0.94 (0.65–1.38)	0.99 (0.67–1.46)
Prostate	25/47	97/190	0.97 (0.49–1.89)	0.89 (0.44–1.80)
Age ≥ 60 years				
Melanoma	17/31	57/96	1.00 (0.47–2.14)	1.10 (0.47–2.56)
Breast	81/104	310/380	0.89 (0.60–1.32)	0.85 (0.56–1.29)
Prostate	80/148	368/523	0.74 (0.52–1.06)	0.75 (0.52–1.09)
No diabetes				
Melanoma	37/76	137/261	0.99 (0.57–1.69)	0.99 (0.56–1.75)
Breast	145/258	545/989	0.93 (0.69–1.24)	0.92 (0.69–1.25)
Prostate	92/172	406/622	0.81 (0.57–1.16)	0.82 (0.57–1.17)
No concomitant use of other drugs used in alcohol dependence[b]				
Melanoma	36/78	143/272	0.93 (0.54–1.59)	0.97 (0.55–1.72)
Breast	144/254	556/990	0.91 (0.68–1.21)	0.90 (0.67–1.20)
Prostate	98/185	441/694	0.79 (0.57–1.09)	0.79 (0.57–1.10)
No conditions related to heavy alcohol abuse[c]				
Melanoma	26/73	100/216	0.74 (0.39–1.41)	0.71 (0.35–1.44)
Breast	94/202	338/816	1.13 (0.78–1.64)	1.16 (0.79–1.70)
Prostate	64/153	291/562	0.71 (0.47–1.08)	0.78 (0.50–1.21)

COPD, chronic obstructive pulmonary disease; ICD, International Classification of Diseases; OR, odds ratio.
[a]Adjusted for COPD; any registered condition related to heavy alcohol abuse; diabetes, Charlson comorbidity index and highest completed education. Furthermore, adjusted for use of 5-α-reductase inhibitors in analyses for prostate cancer, hormone supplements in analyses for breast cancer, and melanoma and thiazides in analyses for melanoma.
[b]Defined as at least one prescription from within ATC-group N07BB (not including disulfiram) >1 year before the index date.
[c]ICD-8, alcoholic psychosis, alcohol dependence syndrome, alcohol-induced chronic pancreatitis; ICD-10, degeneration of nervous system because of alcohol, alcoholic polyneuropathy, alcoholic myopathy, alcoholic cardiomyopathy, alcohol dependence, alcoholic liver disease, alcohol-induced chronic pancreatitis, problematic alcohol consumption.

Clinical studies on the potential antineoplastic effect of disulfiram are sparse. In a randomized-controlled trial, Dufour *et al.* (1993) reported a lower mortality in patients with breast cancer when the disulfiram metabolite dithiocarb was added to cisplatin. Disulfiram treatment was applied for 9 months and was generally well tolerated. Verma *et al.* (1990) could not replicate these findings convincingly in a subsequent trial evaluating the effect of disulfiram added to cisplatin among 53 patients with different types of cancer. However, only 30 patients were eligible for evaluation and the results indicated a trend toward better survival in the disulfiram group.

Laboratory studies have also pointed to a possible antineoplastic effect of disulfiram against glioblastoma and hematological malignancies (Conticello *et al.*, 2012; Liu *et al.*, 2012). As these cancer types occur less frequently in Denmark than melanoma, breast, and prostate cancer, we did not have sufficient power to investigate these suggestions further. Future studies should address whether disulfiram has broader antineoplastic effects. Clinical studies are currently evaluating whether disulfiram plays a role as adjuvant therapy for several cancer types. As our results point to a possible cancer-preventive effect of disulfiram, it will be interesting to observe whether an antineoplastic effect of disulfiram can be extended to the clinical setting as well.

Conclusion

We found a slight, nonsignificant reduction in the risk of breast and prostate cancer with long-term use of disulfiram. Dose–response trends tended to support the finding for prostate cancer. Although we were able to include cancer diagnoses for the entire population of Denmark for a 10-year period, our study had limited statistical precision. Future studies using multiple administrative databases may provide more definite results on the association between disulfiram and the risk of cancer.

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Conflicts of interest

There are no conflicts of interest.

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