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Diabetes mellitus, metformin and head and neck cancer

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SUMMARY

Introduction: Diabetes mellitus (DM (Diabetes Mellitus)) is directly associated with some cancers. However, studies on the association between diabetes mellitus and head and neck cancer (HNC (Head and Neck Cancer)) have rendered controversial results. The objective of this study was to evaluate the association between DM and HNC, as well as the impact of metformin use on the risk of HNC.

Material and methods: This case-control study was conducted within the framework of the Brazilian Head and Neck Genome Project in 2011–2014. The study included 1021 HNC cases with histologically confirmed squamous cell carcinoma of the head and neck admitted to five large hospitals in São Paulo state. A total of 1063 controls were selected in the same hospitals. Odds ratios (OR) and 95% confidence intervals (CI) were estimated using unconditional logistic regression.

Results: Diabetic participants had a decreased risk of HNC (OR = 0.68; 95% CI: 0.49–0.95) than non-diabetic participants, and this risk was further decreased among diabetic metformin users (OR = 0.54; 95% CI: 0.29–0.99). Diabetic metformin users that were current smokers (OR = 0.13; 95% CI: 0.04–0.44) or had an alcohol consumption of >40 g/day (OR = 0.31; 95% CI: 0.11–0.88) had lower risk of HNC than equivalent non-diabetic participants.

Conclusion: The risk of HNC was decreased among diabetic participants; metformin use may at least partially explain this inverse association.

Introduction

Head and neck cancer (HNC) includes tumours of the oral cavity, oropharynx, hypopharynx, and larynx. Nasopharyngeal cancer is also a HNC sub-site but is usually considered a separate disease with a distinct aetiology and particular characteristics [1]. Approximately 600,000 cases of HNC are diagnosed each year, and HNC accounts for 4% of cancer mortality worldwide [2]. More than 90% of HNC are squamous cell carcinoma [3]. The main risk factors associated with HNC are smoking and alcohol consumption, and the interaction between these factors can increase the risk of HNC [4]. Other risk factors include poor oral health, diet, genetic factors, low body mass index (BMI), and occupational factors [5–8].

The association between diabetes mellitus (DM) and the increased risk of certain cancers, such as liver, pancreatic, colon, kidney, bladder, endometrial, and breast cancer, is well established [9–11], while the risk of prostate cancer is decreased among diabetic patients [11,12]. Although some studies with DM has also

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Table 1
Description of excluded and included cases and controls.

Description	Subjects
Cases included	1021
Cases excluded	270
Missing anatomical location of tumor	126
Missing morphology information	50
Cancer diagnosis not confirmed	34
Cancer at other sites (not HNC)	21
Others histology (not SCC)	12
Cancer in situ	9
Synchronous cancers	7
Previous cancer treatment	4
Cancers with previous HNC treatment	3
Repeat cases	3
Missing information on diabetes mellitus	1
Total cases	1291
Controls included	1063
Controls excluded	52
Did not meet inclusion criteria	36
With cancer diagnosis	13
Repeated controls	3
Total controls	1115

HNC: head and neck cancer; SCC: squamous cell carcinoma.

been associated with HNC, these results are still controversial. In some studies, diabetic patients had an increased risk of cancer at some HNC sub-sites [10,13–15], while in other studies this risk was decreased [9,16].

One possible explanation for inverse association between DM and some kinds of cancers is metformin use among diabetic patients. Metformin is a medication used to control Type 2 DM and can inhibit cell proliferation, which has been inversely associated with cancer risk [11,17]. It has been shown that metformin users have a reduced risk of colorectal, liver, lung, and prostate cancer [18–20]. Studies on the association between DM and the risk of HNC that take into account metformin use have also reported conflicting results. A Taiwanese study reported a decreased risk of HNC among metformin users (adjusted hazard ratio = 0.66; 95% confidence interval [CI] 0.55–0.79) [21], while another study in the United Kingdom reported no association [22].

The objective of this study was to evaluate the association between DM and HNC, as well as the impact of metformin use on the risk of HNC.

Material and methods

This case-control study was conducted within the framework of the Brazilian Head and Neck Genome Project (GENCAPO) from December 2011 to November 2014. The study recruited 1291 HNC cases admitted to three general hospitals and two cancer hospitals in Sao Paulo state, Brazil. All HNC cases had histologically confirmed squamous cell carcinoma of the head and neck, and the International Classification of Diseases, 10th Revision [23] was used to classify these cancers into five sub-sites [24]: oral cavity, oropharynx, hypopharynx, larynx, and oral-oropharynx-hypopharynx not specified.

A total of 1115 controls were selected from the same hospitals as the HNC cases: in the general hospitals we recruited hospital controls who were individuals admitted with diseases other than cancer (for example, diseases related to skin, eyes, ears, genitourinary tract, circulatory disorders, nervous system disorders and others); visitor controls were recruited in the cancer hospitals, excluding visitors of HNC patients, since they could have similar habits to the HNC patients.

After exclusions, the final study sample comprised 1021 cases and 1063 controls (Table 1). Controls were frequency-matched to HNC cases by sex and age (in 5-year groups). This study was approved by the institutional ethical review boards of all hospitals, and all participants gave written informed consent.

Data collection

Participants were interviewed using a standardised questionnaire to collect information on socio-demographic, socio-economic, and lifestyle factors, as well as family history of cancer. Authors extracted information on age, sex, education, BMI (based on height and weight 2 years before the interview), DM, tobacco consumption, and alcohol consumption from the questionnaire.

Blood samples were also collected from cases and controls, stored in tubes containing EDTA at −70 °C, and used for glycated haemoglobin tests (A1C). A1C provides an average assessment of glucose control for the previous 60–90 days without the need for fasting. A1C was performed using 1 ml of blood at a laboratory certified by the National Glycohemoglobin Standardization Program and was considered positive for diabetes when values were above 6.5% [25].

The final DM variable was constructed by combining information from three sources in order to ensure a better characterisation of the main explanatory variable and avoid underestimating the prevalence of DM. Thus, participants who reported they were diabetic at interview, or had a diagnosis of DM in medical records, or had a positive A1C result were categorised as diabetic (Table 2). Patients with self-reported DM were categorised as diabetic even if A1C values were below 6.5% (controlled DM). Diabetic participants were then further categorised as metformin users and non-users. Information about metformin use was taken from medical records.

Tobacco consumption was assessed in pack-years, calculated by multiplying the average number of packs of cigarettes smoked in 1 day by the number of years the participant smoked. One cigarette, one pipe and one cigar are equivalent to 1 g, 3.5 g and 4 g of tobacco, respectively. Analyses were performed for both smoking status and pack-years of tobacco consumption.

Alcohol consumption was assessed in g/day. One litter of different alcoholic beverages was converted to 5%, 12%, and 40% of alcohol for beer, wine, and spirits, respectively. Consumption was then converted to grams of alcohol (one litter is equivalent to 798 g of alcohol) and the daily average alcohol intake during the consumption period was calculated. Analyses were performed for both drinking status and g/day of alcohol consumption.

Statistical analysis

Odds ratios (OR) and 95% CI were estimated using unconditional logistic regression. Analyses comparing participants with and without DM were adjusted for sex, age, education, BMI (in kg/m^2), tobacco consumption, and alcohol consumption. The models were also adjusted for hospital of recruitment (centre), since heterogeneity was detected by the likelihood ratio-test.

In metformin analyses, diabetic metformin users and non-users were compared with those without DM. However, these analyses did not include controls who were visitors in cancer hospitals. Indeed, as they were not patients they did not have medical records available in which to verify metformin use. Consequently, metformin analyses were not adjusted for centre.

Analyses were stratified by sex, HNC sub-site, smoking status, drinking status, tobacco consumption (non-smokers, >0–40, >40 pack-years) and alcohol consumption (non-drinkers, >0–40, >40 g/day).

Missing data were found for the following variables: education (18 cases and two controls), BMI (86 cases and five controls), alcohol consumption (50 cases and 20 controls), and tobacco consump-

tion (seven cases and six controls). Multiple imputation with the PROC MI procedure in version 9.4 of SAS statistical software (SAS Institute, Inc., Cary, North Carolina) was applied in order to include participants with missing data in the analyses. Estimated power for the study was 86.8%. For analysis using metformin information, calculated power was 77.1%. All statistical analyses were conducted using SAS version 9.4 statistical software and a 5% statistical significance level was adopted.

Results

Among the 1021 cases and 1063 controls, a higher percentage of current-smokers were observed in cases (68.0%) compared to controls (16.3%). Of cases, 53.6% were current-drinkers compared with 43.5% of controls. There were 359 participants with DM, 150 (14.7%) cases and 209 (19.7%) controls. A large percentage (18.7%) of diabetic participants was identified through A1C only.

DM was inversely associated with HNC in both males and females and in all HNC sub-sites (Table 3). However, the risk reduction was statistically significant only in males (OR = 0.68; 95% CI: 0.47–0.99) and in pharyngeal cancer (OR = 0.43; 95% CI: 0.27–0.68). The decreased risk of HNC associated with DM was most remarkable among heavy smokers (>40 pack-years) and heavy drinkers (>40 g/day) (Table 4).

In analyses considering use of metformin were included 1021 cases and 587 hospital controls, only those in which it was possible to confirm or not the use of metformin. So visitor controls who did not have metformin information were excluded from the analysis. Metformin use among diabetic participants was associated with a decreased risk of HNC. A negative association was also observed in patients with pharyngeal and laryngeal cancer (Table 5), as well as

Table 2
Selected characteristics of the study sample by case/control status.

		Cases		Controls	
		n	%	n	%
Sex	Male	884	86.6%	693	65.2%
	Female	137	13.4%	370	34.8%
Education	No formal education	136	13.6%	74	7.0%
	Incomplete junior high school	581	57.9%	473	44.6%
	Junior high school	137	13.7%	162	15.3%
	High school graduate	119	11.9%	239	22.5%
	College graduate	30	3.0%	113	10.7%
BMI	<18.5	48	5.1%	25	2.4%
	18.5 <25	536	57.3%	379	36.3%
	25 <30	252	27.0%	375	35.9%
	⩾30	99	10.6%	266	25.5%
Smoking status	Non-smoker	76	7.5%	525	49.7%
	Former smoker	249	24.5%	360	34.1%
	Current smoker	690	68.0%	172	16.3%
Tobacco consumption (pack-years)	Non-smoker	76	7.5%	525	49.7%
	>0–10	61	6.0%	196	18.5%
	11–20	65	6.4%	95	9.0%
	21–30	104	10.2%	63	6.0%
	31–40	128	12.6%	57	5.4%
	41–50	135	13.3%	44	4.2%
	⩾51	453	44.6%	106	10.0%
Alcohol drinking status	Non-drinker	84	8.7%	353	33.8%
	Former drinker	367	37.8%	236	22.6%
	Current drinker	520	53.6%	454	43.5%
Alcohol consumption (g/day)	Non-drinker	84	8.7%	353	33.8%
	>0–10	87	9.0%	291	27.9%
	>10–20	52	5.4%	121	11.6%
	>20–30	46	4.7%	63	6.0%
	>30–40	51	5.3%	45	4.3%
	>40–50	46	4.7%	29	2.8%
	>50	605	62.3%	141	13.5%
Type of hospital	General hospital	429	42.0%	587	55.2%
	Cancer hospital	592	57.9%	476	44.8%
Presence of DM	No	871	85.3%	854	80.3%
	Yes	150	14.7%	209	19.7%
HNC sub-site	Oral cavity	349	34.2%		
	Oropharynx	298	29.2%		
	Hypopharynx	92	9.0%		
	Larynx	214	21.0%		
	HNC without specification	40	3.9%		
	Multiple localizations	28	2.7%		
		n	Mean (SD)	n	Mean (SD)
Age		1021	59 (10.2)	1063	56.3 (12.9)
Weight		980	68.7 (14.1)	1058	75.2 (16.6)

BMI: body mass index; DM: diabetes mellitus; HNC: head and neck cancer.

Table 3
Odds ratios (OR) and confidence intervals (CI) of the association between diabetes mellitus (DM) and risk of head and neck cancer (HNC) for all participants and stratified by sex and HNC sub-site.

		Cases		Controls		p-value	OR	95% CI
		n	%	n	%			
All individuals[a]		(n = 1021)		(n = 1063)				
	Without DM	871	85.3%	854	80.3%	0.022	1	
	With DM	150	14.7%	209	19.7%		0.68	(0.49–0.95)
By sex[b]								
Male		(n = 884)		(n = 693)				
	Without DM	758	85.7%	554	79.9%	0.042	1	
	With DM	126	14.3%	139	20.1%		0.68	(0.47–0.99)
Female		(n = 137)		(n = 370)				
	Without DM	113	82.5%	300	81.1%	0.532	1	
	With DM	24	17.5%	70	18.9%		0.80	(0.39–1.63)
By HNC sub-site[a]								
Oral cavity		(n = 349)		(n = 1063)				
	Without DM	288	82.5%	854	80.3%	0.221	1	
	With DM	61	17.5%	209	19.7%		0.75	(0.48–1.19)
Pharynx (oropharynx + hypopharynx)		(n = 390)		(n = 1063)				
	Without DM	345	88.5%	854	80.3%	<0.001	1	
	With DM	45	11.5%	209	19.7%		0.43	(0.27–0.68)
Larynx		(n = 214)		(n = 1063)				
	Without DM	175	81.8%	854	80.3%	0.465	1	
	With DM	39	18.2%	209	19.7%		0.83	(0.51–1.37)

[a] OR adjusted for age, sex, body mass index, tobacco, alcohol and centre.
[b] OR adjusted for age, body mass index, tobacco, alcohol and centre.

among current smokers (OR = 0.13; 95% CI: 0.04–0.44) and heavy drinkers (OR = 0.31; 95% CI: 0.11–0.88) (Table 6).

Although there is no sufficient statistical power to perform separate analysis for oropharyngeal and hypopharyngeal cancer, an exploratory analysis indicated a lower risk of hypopharyngeal cancer in diabetic metformin users. The risk of oropharyngeal cancer was decreased in both metformin users and non-users (data not shown).

Discussion

In this case-control study was found an inverse association between DM and HNC. This association was also observed in subgroups such as men, current smokers, heavy drinkers, and participants with pharyngeal and laryngeal cancer. A possible explanation for this decreased risk could be metformin use in participants with DM. Indeed, metformin users in our study had the lowest risk of HNC, and a decreased risk of HNC was also observed in metformin users who were current smokers or heavy drinkers.

This study has several strengths, including the availability of A1C results for most participants. Indeed, if authors had used self-reported information only, as has been done in other studies, a considerable percentage of cases (30.5%) and controls (28%) would have been wrongly classified as not having DM, and the observed association between DM and HNC would have been biased. However, the study also has limitations. Controls who were visitors in cancer hospitals were excluded from metformin analyses, since they did not have medical records in which were possible verify metformin use. Moreover, as in other studies, we were unable to separate DM by type (Type 1 or Type 2), as this information was missing for 33.3% of cases and 25.8% of controls. Although metformin is mostly used in Type 2 DM, metformin use in individuals with Type 1 DM has increased in recent years, as it may lead to contribute to a decrease in the daily dose of insulin [26]. In present study, metformin was used by five (27.8%) of the 18 participants with Type 1 DM. Although it is important, as in the most studies evaluating the association between metformin, DM and cancer, the present study could not be adjusted for dosage or duration of metformin use since this information was present only for 5% of DM individuals.

Previous studies on the association between DM and HNC subsites have shown contradictory results [9,10,13–16,27]. Findings in the current study were similar to those from the SEER database for people over 68 years old (OR = 0.92; 95% CI: 0.88–0.96) [16] and those of a study on men (OR = 0.85 for cancer of the oral cavity + pharyngeal cancer, 95% CI: 0.82–0.89; OR = 0.76 for laryngeal cancer, 95% CI: 0.71–0.80) [9]. In agreement with current study, some previous studies have reported a decreased cancer risk among metformin users. However, the protective effect of metformin use on HNC is not conclusive [21,22]. Sikka et al. [28] demonstrated that metformin may inhibit cell growth in squamous cell carcinoma of the head and neck. Thus the protection conferred by metformin use may be explained by its role in the activation of adenosine monophosphate-activated protein kinase (AMPK) [19,29]. AMPK is an enzyme and its activation inhibits protein synthesis and the action of the mTOR protein. mTOR is important for cell growth and its activity is deregulated in various cancers [19,30]. The mTOR pathway is regulated by both AMPK and AKT, which have opposite functions. In response to an external stimulus, AKT activates the mTOR pathway, contributing to cell proliferation. On the other hand, the AMPK can reduce the action of mTOR. Metformin use may activate AMPK, thus inhibiting mTOR action and decreasing cell proliferation [11,19,31].

Persistent activation of AKT is present in various cancers, including HNC [32]. Although tobacco and alcohol consumption are the main risk factors for HNC, the molecular mechanism involved is not clear and few studies have investigated the association between these risk factors and AKT and mTOR activation, especially in HNC. One study showed that alcohol and tobacco activate AKT [33]. Another study showed that NKK, a main tobacco carcinogen, is associated with activation of AKT in HNC patients [34]. Likewise, tobacco promotes lung tumorigenesis through activation of signalling the AKT/mTOR pathway [35].

Present results indicated that DM was inversely associated with HNC in current smokers and heavy smokers, though this inverse association was restricted to metformin users. The present study

Table 4
Odds ratios (OR) and confidence intervals (CI) of head and neck cancer (HNC) according to diabetes mellitus (DM) by level of tobacco and alcohol consumption.

		Cases		Controls		p-value	OR[a]	95% CI
		n	%	n	%			
Smoking status[a]								
Non-smoker		(n = 75)		(n = 525)				
	Without DM	62	82.7%	433	82.5%	0.926	1	
	With DM	13	17.3%	92	17.5%		1.04	(0.50–2.16)
Former smoker		(n = 249)		(n = 360)				
	Without DM	187	75.1%	272	75.6%	0.966	1	
	With DM	62	24.9%	88	24.4%		1.01	(0.63–1.62)
Current smoker		(n = 690)		(n = 172)				
	Without DM	619	89.7%	145	84.3%	0.003	1	
	With DM	71	10.3%	27	15.7%		0.42	(0.24–0.74)
Drinking status[b]								
Non-drinker		(n = 84)		(n = 353)				
	Without DM	70	83.3%	278	78.8%	0.086	1	
	With DM	14	16.7%	75	21.2%		0.47	(0.20–1.11)
Former drinker		(n = 367)		(n = 236)				
	Without DM	309	84.2%	185	78.4%	0.927	1	
	With DM	58	15.8%	51	21.6%		1.03	(0.58–1.82)
Current drinker		(n = 520)		(n = 454)				
	Without DM	448	86.2%	374	82.4%	0.159	1	
	With DM	72	13.8%	80	17.6%		0.71	(0.45–1.14)
Tobacco consumption (in pack-years)[a]								
Non-smoker		(n = 75)		(n = 525)				
	Without DM	62	82.7%	433	82.5%	0.926	1	
	With DM	13	17.3%	92	17.5%		1.04	(0.50–2.16)
0–40		(n = 351)		(n = 382)				
	Without DM	303	86.3%	306	80.1%	0.400	1	
	With DM	48	13.7%	76	19.9%		0.81	(0.50–1.32)
>40		(n = 588)		(n = 150)				
	Without DM	503	85.5%	111	74.0%	0.002	1	
	With DM	85	14.5%	39	26.0%		0.44	(0.26–0.74)
Alcohol consumption (in g/day)[b]								
Non-drinker		(n = 84)		(n = 353)				
	Without DM	70	83.3%	278	78.8%	0.086	1	
	With DM	14	16.7%	75	21.2%		0.47	(0.20–1.11)
0–40		(n = 236)		(n = 520)				
	Without DM	194	82.2%	427	82.1%	0.775	1	
	With DM	42	17.8%	93	17.9%		0.93	(0.55–1.57)
>40		(n = 651)		(n = 170)				
	Without DM	563	86.5%	132	77.6%	0.027	1	
	With DM	88	13.5%	38	22.4%		0.56	(0.33–0.94)

[a] OR adjusted for age, sex, body mass index, alcohol and centre.
[b] OR adjusted for age, sex, body mass index, tobacco and centre.

is the first to report a lower risk of HNC among diabetic metformin users who were heavy smokers. Metformin use has been reported to prevent tobacco carcinogen-induced lung tumorigenesis in animal models [36]. Indeed, metformin has been suggested as a chemopreventive agent against lung cancer among heavy smokers [37]. The mechanism of metformin may be similar in HNC and lung cancer, since the main risk factor for both is tobacco smoke.

Our study is the first to investigate the association between metformin use and HNC in diabetic individuals who are heavy drinkers. A decreased risk of HNC is not entirely surprising considering the literature on animal models. In one study, a group of rats with chronic ethanol consumption showed a reduction in AMPK enzyme, but the group with ethanol consumption and treated with metformin showed improvement in hepatic injury through activation of AMPK. The AMPK level was similar in rats without metformin and without alcohol consumption [38]. Among heavy alcohol drinkers, the protective effect for HNC observed in diabetic metformin users might be explained by activation of AMPK by metformin, but further studies should carried out to evaluate and understand this mechanism.

We observed an inverse association between HNC and DM in all metformin users, but the associations were only statistically significant for laryngeal cancer. Becker et al. [22] also reported a reduction in the risk of laryngeal cancer in long-term metformin users. Results also showed an inverse association between pharyngeal cancer and DM, regardless of metformin use. Despite the lack of statistical power to study oropharyngeal and hypopharyngeal cancer separately, the results indicated a reduced risk of hypopharyngeal cancer among diabetic metformin users. All diabetic participants had a reduced risk of oropharyngeal cancer regardless of metformin use. In a pooled analysis, no association was found between DM and cancer of the oropharynx or other HNC subsites [27]. It is noteworthy that pharyngeal cancers are different from laryngeal cancers in pathogenesis and prognosis [3,39], which can explain differences in results. Although the incidence of both oropharyngeal and laryngeal cancer are higher in men, there is a

Table 5
Odds ratios (OR) and confidence intervals (CI) of head and neck cancer (HNC) according to diabetes mellitus (DM) and metformin use for all participants and stratified by sex and HNC sub-site.

		Cases		Controls		p-value	OR	95% CI
		n	%	n	%			
All individuals[a]		(n = 1021)		(n = 587)				
	Without DM	871	85.3%	470	80.1%	0.067	1	
	DM and metformin	40	3.9%	48	8.2%	0.049	0.54	(0.29–0.99)
	DM without metformin	110	10.8%	69	11.8%	0.142	0.72	(0.46–1.12)
By sex[b]								
Male		(n = 885)		(n = 322)				
	Without DM	758	85.7%	257	79.8%	0.089	1	
	DM and metformin	33	3.7%	25	7.8%	0.073	0.51	(0.24–1.07)
	DM without metformin	93	10.5%	40	12.4%	0.134	0.67	(0.39–1.13)
Female		(n = 137)		(n = 265)				
	Without DM	113	82.5%	213	80.4%	0.828	1	
	DM and metformin	7	5.1%	23	8.7%	0.577	0.74	(0.26–2.11)
	DM without metformin	17	12.4%	29	10.9%	0.744	0.88	(0.40–1.91)
HNC sub-site[a]								
Oral cavity		(n = 349)		(n = 587)				
	Without DM	288	82.5%	470	80.1%	0.429	1	
	DM and metformin	22	6.3%	48	8.2%	0.365	0.72	(0.36–1.46)
	DM without metformin	39	11.2%	69	11.8%	0.293	0.74	(0.42–1.30)
Pharynx (oropharynx + hypopharynx)		(n = 390)		(n = 587)				
	Without DM	345	88.5%	470	80.1%	0.002	1	
	DM and metformin	11	2.8%	48	8.2%	0.023	0.34	(0.13–0.86)
	DM without metformin	34	8.7%	69	11.8%	0.004	0.41	(0.22–0.75)
Larynx		(n = 214)		(n = 587)				
	Without DM	175	81.8%	470	80.1%	0.049	1	
	DM and metformin	7	3.3%	48	8.2%	0.019	0.28	(0.10–0.81)
	DM without metformin	32	15.0%	69	11.8%	0.655	1.16	(0.60–2.26)

[a] OR adjusted for age, sex, body mass index, tobacco and alcohol.
[b] OR adjusted for age, body mass index, tobacco and alcohol.

Table 6
Odds ratios (OR) and confidence intervals (CI) of head and neck cancer (HNC) according to diabetes mellitus (DM) and metformin use by smoking and drinking status and consumption level of tobacco and alcohol.

		Cases		Controls		p-value	OR	95% CI
		n	%	n	%			
Smoking status[a]								
Non smoker		(n = 75)		(n = 307)				
	Without DM	62	82.7%	246	80.1%	0.421	1	
	DM and metformin	6	8.0%	29	9.4%	0.443	0.68	(0.25–1.83)
	DM without metformin	7	9.3%	32	10.4%	0.250	0.57	(0.22–1.49)
Former smoker		(n = 249)		(n = 178)				
	Without DM	187	75.1%	138	77.5%	0.903	1	
	DM and metformin	20	8.0%	14	7.9%	0.854	0.92	(0.40–2.14)
	DM without metformin	42	16.9%	26	14.6%	0.711	1.13	(0.59–2.15)
Current smoker		(n = 690)		(n = 197)				
	Without DM	619	89.7%	83	85.6%	0.003	1	
	DM and metformin	13	1.9%	5	5.2%	0.001	0.13	(0.04–0.44)
	DM without metformin	58	8.4%	9	9.3%	0.269	0.61	(0.26–1.46)
Drinking status[b]								
Non drinker		(n = 84)		(n = 238)				
	Without DM	70	83.3%	185	77.7%	0.118	1	
	DM and metformin	5	6.0%	25	10.5%	0.274	0.53	(0.17–1.66)
	DM without metformin	9	10.7%	28	11.8%	0.061	0.39	(0.15–1.05)
Former drinker		(n = 367)		(n = 143)				
	Without DM	309	84.2%	112	78.3%	0.818	1	
	DM and metformin	14	3.8%	11	7.7%	0.541	0.72	(0.26–2.05)
	DM without metformin	44	12.0%	20	14.0%	0.934	1.03	(0.49–2.19)
Current drinker		(n = 520)		(n = 189)				
	Without DM	448	86.2%	159	84.1%	0.841	1	
	DM and metformin	21	4.0%	12	6.3%	0.572	0.74	(0.27–2.08)
	DM without metformin	51	9.8%	18	9.5%	0.818	0.91	(0.41–2.02)

Table 6 (*continued*)

		Cases		Controls		p-value	OR	95% CI
		n	%	n	%			
Tobacco consumption (in pack-years)[a]								
Non-smoker		(n = 75)		(n = 307)				
	Without DM	62	82.7%	246	80.1%	0.421	1	
	DM and metformin	6	8.0%	29	9.4%	0.443	0.68	(0.25–1.83)
	DM without metformin	7	9.3%	32	10.4%	0.250	0.57	(0.22–1.49)
0–40		(n = 351)		(n = 209)				
	Without DM	303	86.3%	172	82.3%	0.164	1	
	DM and metformin	13	3.7%	13	6.2%	0.065	0.42	(0.17–1.06)
	DM without metformin	35	10.0%	24	11.5%	0.816	1.08	(0.56–2.10)
>40		(n = 588)		(n = 66)				
	Without DM	503	85.5%	49	74.2%	0.718	1	
	DM and metformin	20	3.4%	6	9.1%	0.066	0.36	(0.12–1.07)
	DM without metformin	65	11.1%	11	16.7%	0.104	0.53	(0.25–1.14)
Alcohol consumption (in g/day)[b]								
Non-drinker		(n = 84)		(n = 238)				
	Without DM	70	83.3%	185	77.7%	0.118	1	
	DM and metformin	5	6.0%	25	10.5%	0.274	0.53	(0.17–1.66)
	DM without metformin	9	10.7%	28	11.8%	0.061	0.39	(0.15–1.05)
0–40		(n = 236)		(n = 248)				
	Without DM	194	82.2%	203	81.9%	0.924	1	
	DM and metformin	15	6.4%	16	6.5%	0.704	1.21	(0.46–3.16)
	DM without metformin	27	11.4%	29	11.7%	0.958	0.98	(0.48–2.00)
>40		(n = 651)		(n = 84)				
	Without DM	563	86.5%	68	81.0%	0.036	1	
	DM and metformin	20	3.1%	7	8.3%	0.028	0.31	(0.11–0.88)
	DM without metformin	68	10.4%	9	10.7%	0.571	0.78	(0.34–1.83)

[a] OR adjusted for age, sex, body mass index and alcohol.
[b] OR adjusted for age, sex, body mass index and tobacco.

slightly higher incidence in oropharyngeal cancer among women in some European countries [2,39,40]. Unlike the laryngeal cancer, an increase in oropharyngeal incidence has occurred in whites and younger individuals [39]. There is growing evidence that a substantial proportion of oropharyngeal cancers are associated with human papillomavirus (HPV) infection [39,40]. Unfortunately, there was no information on HPV in our study; therefore this information was not added in the models, which could have caused some bias in the results for oropharyngeal cancer.

Conclusion

We found an inverse association between DM and HNC. The protective effect of DM was at least partially explained by metformin use, since metformin users had an even lower risk of HNC. The inverse association of HNC risk among metformin users was stronger in current smokers and heavy drinkers.

Conflict of interest

None declared.

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