Tampere University

MARYAM HADJI

Opium Use and Risk of Cancer

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ABSTRACT

The use of opium has been classified by the International Agency for Research on Cancer (IARC) as a carcinogenic to humans (Group 1 carcinogen), but there is still a lot that needs to be clarified regarding the association between opium use and cancer. To study this association further, I used the data collected by the Iranian Opium and Cancer (IROPICAN) study in ten provinces of Iran. This study focuses on examining the association between opium use and cancers of the head and neck, bladder, colon, and rectum. The overall aim of this project is to evaluate the association between the consumption of opium and cancers of the head and neck, colorectum, and bladder.

This dissertation utilized 633 histologically confirmed cancer cases of head and neck squamous cell carcinoma (HNSCC), 717 cases of bladder cancer (BC), 848 cases of colorectal cancer (CRC), and 3,477 common controls selected from hospital visitors who did not have cancer and were not relatives or friends of the cancer cases. The extensive questionnaire used in the study covered a variety of topics, including opium use (e.g., information on age at initiation, duration, frequency, typical amount, and route), as well as potential confounding factors such as tobacco use (e.g., cigarettes, nass, and waterpipe), and dietary factors. The response rate was 99% for the cancer cases and 89% for the controls.

In the validation and pilot phases of the study, the reported opium use showed a reasonably good level of accuracy, with a sensitivity of 70% for cancer cases and 69% for controls. The study showed excellent reliability with intra-class correlation coefficients of 0.96 for ever opium use and 0.88 for regular opium use.

Multivariable unconditional logistic regression models were used to estimate the odds ratios (OR) and 95% confidence intervals (CI). The ORs were adjusted for potential confounders for each cancer type. Regular opium use was associated with a highly increased risk of HNSCC (OR: 3.8, 95% CI: 3.0, 4.8). There was a strong dose–response relationship between opium use and HNSCC risk, with increasing risk seen with higher frequency and amount of use, and with longer duration. Regular opium use was found to significantly increase the risk of cancers of the pharynx OR 2.9, 95% CI: 1.4, 6.0), larynx (OR 6.6, 95% CI: 4.7, 9.1), and other sites within the head and neck region (OR: 6.0, 95% CI: 2.4, 14.7). There was a significant interaction between opium use and cigarette smoking affecting HNSCC risk, with an OR of 8.2 (95% CI: 6.2, 10.7) among those who used opium and smoked cigarettes at the same time compared to those who had never used anything, but the association with opium remained significant among individuals who had never used tobacco.

Regular opium consumption was also associated with an increased risk of BC, with an OR of 3.5 (95% CI: 2.8, 4.3) compared to those who had never used opium. The risk of BC decreased to one-third among individuals who had stopped using opium more than ten years before the date of the interview, compared to those who still used opium. The risk of developing BC was found to be 4.8 (95% CI: 3.7, 6.3) among current opium users. However, for those who had stopped using opium more than 10 years prior to the interview, the risk of BC decreased to 1.5 (95% CI: 1.0, 2.4). The OR for individuals who used both crude opium (teriak) and opium juice was 7.4 (95% CI: 4.1, 13.3). There was a joint effect of opium and tobacco, with an OR of 7.7 (95% CI: 6.0, 9.7) for individuals who used both opium and tobacco.

No association was found between regular opium consumption and the risk of CRC (OR 0.9, 95% CI: 0.7, 1.2) compared to individuals who never used opium. Still, the study suggests that opium use two or more times per day may be associated with an increased risk of CRC with an OR of 2.0 (95% CI: 1.1, 3.8) compared to non-users of opium.

Regular opium use was found to be strongly associated with an increased risk of developing BC and HNSCC, but the link between regular opium use and CRC was weak. The studies included in my thesis will add to the understanding of the impact of opium use on the risk of developing cancer in the head and neck, bladder, colon, and rectum, and serve as a crucial background for future investigations in this field.

TIIVISTELMÄ

Kansainvälinen syöväntutkimuskeskus (International Agency for Research on Cancer, IARC) arvioi hiljattain oopiumin käytön syövän vaaraa lisääväksi ykkösluokan karsinogeeniksi, mutta tieto oopiumin käytön yhteydestä syöpäsairauksiin on yhä puutteellista. Hyödynsin väitöskirjassani Iranian Opium and Cancer (IROPICAN) -tutkimushankkeessa kymmenessä Iranin maakunnassa kerättyä tietoaineistoa. Tutkimukseni käsittelee pään ja kaulan alueen, virtsarakon ja suoliston syöpien yhteyttä oopiumin käyttöön.

Tutkimukseni perustuu 633 histologisesti varmennettuun pään ja kaulan alueen levyepiteelikarsinoomaan, 717 virtsarakon syöpään ja 848 suolistosyöpään sairastuneen henkilön ja kaikille syöpälajeille yhteisten 3477 verrokkihenkilön tietoihin. Verrokkihenkilöt olivat sairaaloissa vierailleita henkilöitä, joilla ei ollut syöpää ja jotka eivät olleet syöpäpotilaiden sukulaisia eivätkä ystäviä. Tiedot kerättiin laajan haastattelulomakkeen avulla. Siinä kysyttiin opiumin käytön aloitusikää, käytön kestoa, tiheyttä, käytetyn opiumin määrää ja käyttötapaa. Lisäksi kysyttiin mahdollisista sekoittavista tekijöistä kuten tupakan, nuuskan ja vesipiipun käytöstä ja ravintotekijöistä. Vastausprosentti oli tutkimukseen valittujen syöpätapausten joukossa 99 % ja verrokkien joukossa 89 %.

Tutkimuksen laadunvarmistus- ja pilottivaiheissa todettiin oopiuminkäyttötietojen raportoinnin olevan suhteelliseen luotettavaa: 70 prosentilla tapauksista ja 69 prosentilla verrokeista itse ilmoitettu tieto siitä, käyttääkö hän oopiumia vai ei, piti paikkansa. Tieto piti paikkansa peräti 96 prosentilla tapauksista ja 88 prosentilla verrokeista, jotka kertoivat käyttävänsä oopiumia säännöllisesti.

Analyyseissä oopiumin vaikutusta kuvattiin ristitulosuhteilla (odds ratio, OR), jotka laskettiin logistisella regressiolla ja korjattiin sekoittajatekijöiden vaikutuksista. Oopiumin säännölliseen käyttöön liittyi suurentunut pään ja kaulan levyepiteelikarsinoomaan riski (OR: 3,8; 95 %:n luottamusväli: 3,0–4,8). Riski kasvoi, kun käyttötiheys ja -määrä lisääntyivät ja kesto piteni. Oopiumin säännöllisen käytön

havaittiin lisäävän eniten kurkunpään syövän riskiä (OR: 6,6; 4,7–9,1). Oopiumin käytöllä ja tupakoinnilla oli merkittävä yhteisvaikutus pään ja kaulan levyepiteelikarsinooman riskiin: OR oli 8,2 (6,2–10,7) niillä, jotka sekä käyttivät oopiumia että polttivat savukkeita, verrattuna niihin, jotka eivät olleet koskaan tehneet kumpaakaan.

Oopiumia säännöllisesti käyttävillä oli 3,5-kertainen (2,8–4,3) virtsarakkosyövän riski verrattuna niihin, jotka eivät olleet koskaan käyttäneet oopiumia. OR oli 4,8 (3,7–6,3) tutkittavilla, jotka haastattelun aikaan käyttivät yhä oopiumia, mutta vain 1,5 (1,0–2,4) niillä, jotka olivat lopettaneet oopiumin käytön yli 10 vuotta ennen haastattelua. Henkilöillä, jotka käyttivät sekä raakaa oopiumia että oopiumimehua, OR oli 7,4 (4,1–13,3). Oopiumin ja tupakan yhteiskäyttö nosti riskin 7,7-kertaiseksi (6,0–9,7).

Oopiumin säännöllisellä käytöllä ei havaittu olevan yleisesti ottaen yhteyttä suolistosyövän riskiin (OR: 0,9; 0,7–1,2). Silti oopiumin käyttö vähintään kaksi kertaa päivässä voi liittyä lisääntyneeseen suolistosyövän riskiin: OR oli 2,0 (1,1–3,8) verrattuna niihin, jotka eivät käytä oopiumia.

Oopiumin säännöllisen käytön havaittiin olevan vahvasti yhteydessä lisääntyneeseen virtsarakkosyövän ja pään ja kaulan alueen levyepiteelikarsinooman riskiin, mutta varsin heikosti tai ei ollenkaan suolistosyövän riskiin. Opinnäytetyöhöni sisältyvät tutkimukset lisäävät ymmärrystä oopiumin käytön vaikutuksesta tutkittujen syöpälajien kehittymisriskiin ja toimivat pontimena alan tuleville tutkimuksille.

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ABBREVIATIONS

AP	Attributed proportion due to interaction
ASR	Age-standardized incidence rate
ASMR	Age-standardized mortality rate
BC	Bladder cancer
BMI	Body mass index
CRC	Colorectal cancer
CI	Confidence interval
cDAG	Causal directed acyclic graph
DMFT	Decayed, missing and filled teeth
EPIC	European Perspective Investigation into Cancer and Nutrition
FFQ	Food frequency questionnaire
FINJEM	Finnish job-exposure matrix
GLOBOCAN	Global cancer
HPV	Human papilloma virus
HNPCC	Hereditary non-polyposis colorectal cancer
HNSCC	Head and neck squamous cell carcinoma
HR	Hazard ratio
IBD	Inflammatory bowel disease
ICC	Intra-class correlation coefficient

ICD-O	International Classification of Disease for Oncology
INHANCE	International head and neck cancer epidemiology
IARC	International Agency for Research on Cancer
INCAS	Iranian National Center for Addiction Studies
IQR	Interquartile range
IROPICAN	Iranian Study of Opium and Cancer
MMT	Methadone maintenance treatment
mRR	Meta–relative risk
NIMAD	National Institute for Medical Research Development (in Iran)
OR	Odds ratio
PAH	Polycyclic aromatic hydrocarbons
PPWL	Perceived physical workload
RERI	Relative excess risk due to interaction
RR	Relative risk
SD	Standard deviation
SEER	Surveillance, Epidemiology, and End Results Program
SES	Socioeconomic status
S	Synergy index
SIR	Standardized incidence ratio
Sn	Sensitivity
Sp	Specificity
TLC	Immunoassay thin layer chromatography
URDS	Urine rapid drug screen
WHO	World Health Organization

ORIGINAL PUBLICATIONS

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INTRODUCTION

Millions of people worldwide, particularly in central Asian countries, consume opium, a highly addictive substance derived from the unripe seedpod of the poppy plant, through illegal means ("World Drug Report" 2019). Opium, which is obtained from the poppy plant, contains alkaloids (such as morphine, codeine, and thebaine) as well as non-alkaloids (such as water, sugars, fat, and meconic acid). It is typically processed by heating, boiling, and drying, and may be mixed with certain chemicals (such as lead or chromium) before it is sold to consumers. Opium can be consumed in its minimally processed forms, including raw opium (teriak), opium sap (shireh), or opium dross (sukhteh) (Amin-Esmaeili et al. 2016). Opium can be ingested or smoked in the forms mentioned above. As a result, it is a complex substance containing numerous chemicals, much like tobacco. Opium is typically subject to minimal processing, involving heating, boiling, drying, and occasionally adding certain chemicals (such as lead or chromium) before it reaches the consumer (Amin-Esmaeili et al. 2016). Medical derivatives of opium, such as opioids, are known to have useful effects, for instance serving as effective analgesics for treating pain in chronic illnesses like cancer. Additionally, some opioids, like codeine, have an anti-cough effect (Adcock 1991; Zeppetella 2011).

Over the last 20 years, case-control and cohort studies have produced significant evidence suggesting that opium use may raise the risk of death in general, as well as mortality related to cardiovascular disease (Nalini et al. 2021) and cancer (Alizadeh et al. 2020; Mousavi et al. 2003). Studies have found that opium use is linked to a greater risk of various types of cancer, including larynx cancer (Alizadeh et al. 2020; Khoo 1981; Mohebbi, Hadji, et al. 2021; Mousavi et al. 2003), bladder cancer (Afshari et al., 2017; Akbari et al., 2015; Sheikh, et al., 2020), lung cancer (MacLennan et al., 1977; Masjedi et al., 2013; Sheikh, et al., 2020), pharynx cancer (Bakhshaee et al. 2017; Fahmy, Sadeghi, and Behmard 1983; Khoo 1981; Mohebbi, Hadji, et al. 2021), stomach cancer (Naghibzadeh et al., 2014; Shakeri et al., 2013; Sheikh, et al., 2020), esophageal cancer (Ghadirian et al., 1985; Malekzadeh et al., 2013; Nasrollahzadeh et al., 2008; Shakeri et al., 2012; Sheikh, et al., 2020), pancreatic cancer (Moossavi et al., 2018; Shakeri et al., 2016; Sheikh, et al., 2020), and colon and rectal cancer (Khosravizadegan et al. 2017; Naghibzadeh-Tahami et al. 2016). Based on the data from these studies, along with additional mechanistic research, the International Agency for Research on Cancer (IARC) has recently classified opium use as a Group 1 carcinogen, which means that it is considered to be carcinogenic in humans (IARC Monographs Vol 126 group 2020).

While the IARC Working Group has determined opium use as a cause of cancers affecting the larynx, lung, and bladder, there are still significant gaps in knowledge regarding the association between opium use and cancer. For instance, further investigation is required to determine the degree of the association with these specific cancers and to study how opium use interacts with tobacco smoking in relation to these cancers. Although some credible data exist, it is not clear whether opium increases the risk of cancers of the esophagus, stomach, or pancreas. Additionally, there are conflicting data regarding some cancers, particularly those affecting the colon and rectum (IARC Monographs Vol 126 group, 2020; Khosravizadegan et al., 2017; Sheikh, et al., 2020). Although epidemiological studies to date suggest that consuming opium in its different forms (teriak, shireh, and sukhteh) and through primary application

routes (ingestion and smoking) may be carcinogenic, further data are needed to confirm these findings. To advance scientific understanding in this area, the Iranian Opium and Cancer (IROPICAN) Study was launched in 2012. This study aims to assess the association between opium exposure and the risk of developing head and neck, bladder, and colon and rectum cancers.

Our aim was to make significant progress in this field by designing studies with much larger sample sizes than any previous studies and gathering comprehensive data on opium use for dose-response analyses, accounting for potential confounding factors and examining potential reverse causality. The findings on HNSCC have contributed to the recent conclusions of the IARC Working Group, particularly in relation to evaluating laryngeal and pharyngeal cancers.

1 LITERATURE REVIEW

1.1 Head and neck squamous cell carcinoma

Head and neck cancer is a term used to describe a group of cancers that arise from the tissues and organs in the head and neck region. According to the American Cancer Society, these cancers include cancers of the oral cavity, pharynx, larynx, nasal cavity, and paranasal sinuses. Among these, head and neck squamous cell carcinoma (HNSCC) is the most common type, accounting for approximately 90% of all cases. Other less common histological subtypes of head and neck cancer include adenocarcinoma, adenoid cystic carcinoma, mucoepidermoid carcinoma, and sarcomas ("American Cancer Society" 2020; Mehanna et al. 2010). The proportion of non-squamous cell carcinoma subtypes varies depending on the location within the head and neck, as well as other factors such as age, sex, and smoking status (Chaturvedi et al. 2013).
HNSCC arises from the mucosal epithelium in various parts of the head and neck, including the oral cavity (such as the lips, buccal mucosa, hard palate, anterior tongue, floor of mouth, and retromolar trigone), nasopharynx, oropharynx (such as the palatine tonsils, lingual tonsils, base of tongue, soft palate, uvula, and posterior pharyngeal wall), hypopharynx (which is the lower part of the throat, extending from the hyoid bone to the cricoid cartilage), and larynx (Figure 1).

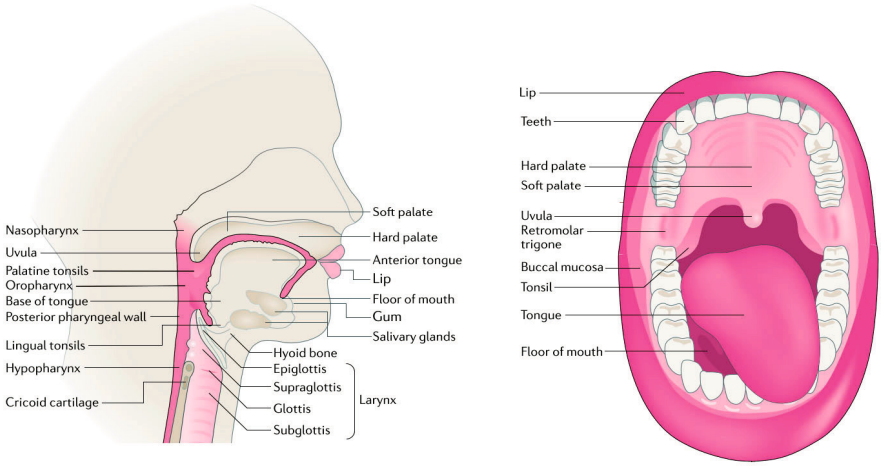

Figure 1. Anatomic sites of head and neck squamous cell carcinoma (Johnson et al. 2020).

The International Classification of Diseases for Oncology (ICD-O) is a system used for coding and classifying neoplasms (abnormal growths of tissue) based on their site (topography) and histology (morphology). It was developed by the World Health Organization (WHO) and is widely used by cancer registries and healthcare providers to accurately classify and report cancer cases. The system allows for consistency and comparability in cancer research, epidemiology, and treatment (Fritz et al. 2019; Canda, Eroğlu, and Hapa 2021). According to the US National Cancer Institute's Surveillance, Epidemiology, and

End Results (SEER) program training module ("National Cancer Institute." 2023), the ICD-O-3 codes used for classifying HNSCC sites comprise several areas in the head and neck, such as the lip, oral cavity (including the tongue, gum, floor of the mouth, palate, and major salivary glands), tonsils, oropharynx, nasopharynx, pyriform sinus, hypopharynx, and larynx (Table 1).

Table 1. ICD-O-3 codes for the head and neck cancer sites (HNSCC).

Site	ICD-O-3
Lip	C00
Base of tongue	C01
Other and unspecified parts of the tongue	C02
Gum	C03
Floor of mouth	C04
Palate	C05
Other and unspecified parts of the mouth	C06
Other and unspecified major salivary gland	C08
Tonsil	C09
Oropharynx	C10
Nasopharynx	C11
Pyriform sinus	C12
Hypopharynx	C13
Other and ill-defined sites in the lip, oral cavity and pharynx	C14
Nasal cavity and middle ear	C30
Accessory sinuses	C31
Larynx	C32

1.1.1 Epidemiology of head and neck squamous cell carcinoma

1.1.1.1 Occurrence

According to data from the Global Cancer Observatory (GLOBOCAN) in 2020, it was estimated that there were more than 878,000 new cases of head and neck cancer including lip, oral cavity, pharynx, and larynx worldwide, resulting in 444,000 reported deaths from these types of cancers ("IARC" 2020). Figures 2–3 depict the age-standardized incidence (ASR) and mortality rates (ASMR) for lip, oral cavity, pharynx, and larynx cancers on a global scale. According to projections from GLOBOCAN, the incidence of these cancers is expected to rise by 30% or approximately 1.08 million new cases annually by the year 2030 (Ferlay et al. 2023).

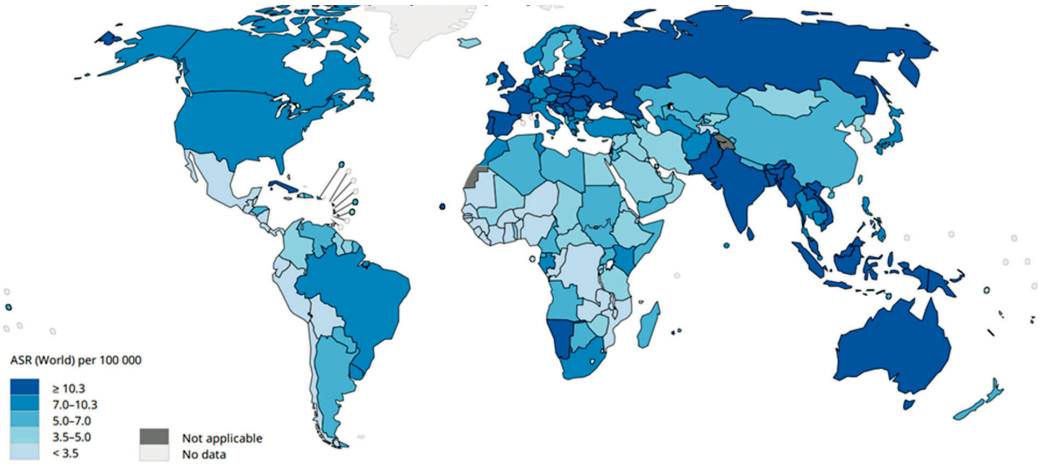

Figure 2. Estimated incidence rates (ASR) of cancers of the lip and oral cavity, pharynx and larynx, by country, age-standardized to the world standard population (GLOBOCAN- 2020) ("IARC" 2020).

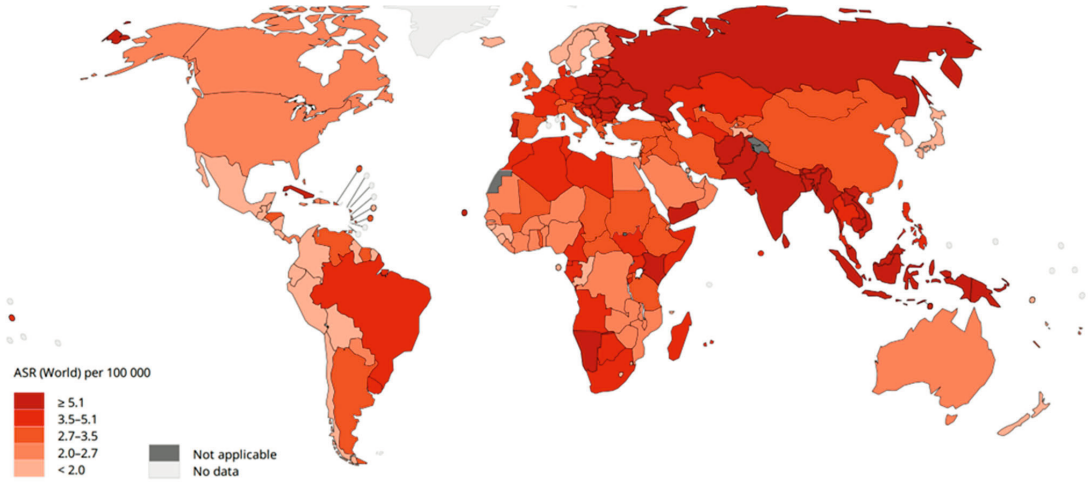

Figure 3. Estimated mortality rates (ASR) of lip, oral cavity, pharynx and larynx cancers, by country, age-standardized to the world standard population (GLOBOCAN- 2020) ("IARC" 2020).

Based on the data from GLOBOCAN 2020, it was estimated that there were approximately 4,097 new cases of cancers affecting the lip, oral cavity, pharynx, and larynx in Iran, resulting in 2,421 deaths attributed to these cancers ("IARC" 2020).

The only published studies on the epidemiology of HNSCC that utilized population-based cancer registry data were carried out in the Golestan province, located in Northern Iran. These studies found that the ASR of HNSCC was 4.8 per 100,000 person-years over a ten-year period from 2004 to 2013 (Taziki et al.

2018). The ASRs for various sites of HNSCC were reported as 1.5 for the oral cavity, 0.3 for the pharynx, and 3.0 for the larynx. The overall ASR for HNSCC was found to be twice as high in Iranian men at 6.6 per 100,000 person-years compared to women at 3.0 per 100,000 person-years. This disparity was primarily attributed to laryngeal cancer (Taziki et al. 2018).

Figure 4 illustrates the ASRs of HNSCC per 100,000 person-years, stratified by sex, over a ten-year period from 2004 to 2013 in the Golestan province and the ASRs of the head and neck cancer cases from 1977 to 2020 in the Nordic countries. The ASR in both areas was consistently higher in men than in women. In the Nordic countries, the ASR has slightly decreased among men whereas it increased among women since the 1970s.

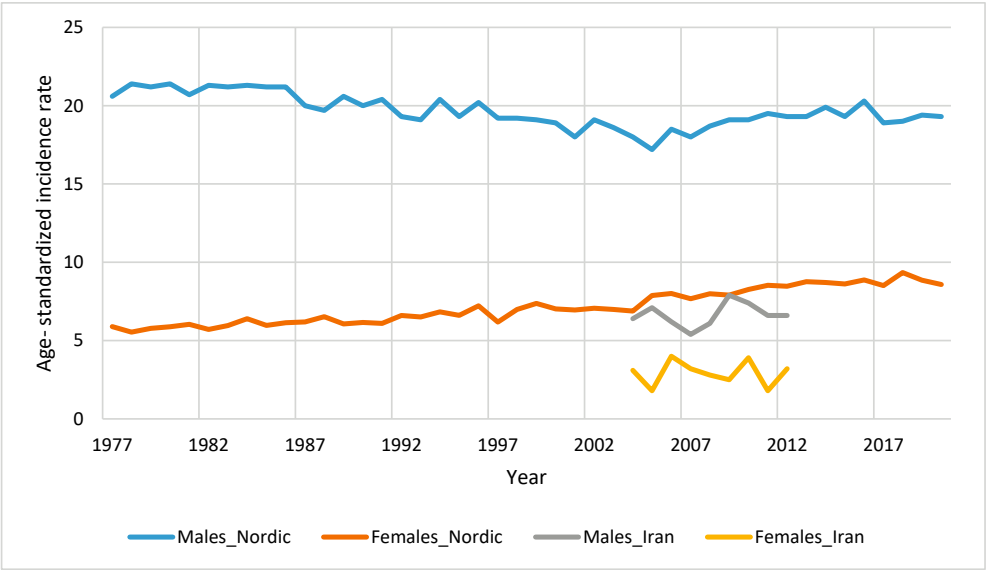

Figure 4. Age-standardized Incidence rates (per 100,000 persons-years) of head and neck squamous cell carcinomas in Golestan Province in Iran (Taziki et al. 2018) and for all head and neck cancers in the Nordic countries during 1977–2020 (*https://Nordcan.Iarc.Fr/En*, 2023), by sex.

1.1.1.2 Etiological factors

The list of risk factors of HNSCC includes tobacco smoking, alcohol consumption, human papillomavirus (HPV), occupation, oral hygiene, hormonal factors in women, lack of physical activity, nutrition, socioeconomic status (SES) (Aupérin 2020; Hashibe et al. 2007; Johnson et al. 2020).

Tobacco smoking

Tobacco smoking is one of the main risk factors for HNSCC, with the strongest risk for laryngeal cancer among those who never drunk alcohol with an odds ratio (OR) of 6.8 and a 95% confidence interval (CI) of 4.3, 11.0 derived from a pooled analysis of 15 case-control studies from Europe, North America, South and Central America (Hashibe et al. 2007). The OR of cigarette smoking for other subsites, including the

oral cavity, was 1.4 (0.9, 2.0), and for the oropharynx and hypopharynx 2.0 (95% CI: 1.3, 3.0) (Hashibe et al. 2007). Di Credico et al. reported the dose-response association between cigarette smoking and HNSCC, using data from 33 case-control studies of the International Head and Neck Cancer Epidemiology (INHANCE) consortium (18,260 cases, 29,844 controls). In this study, they showed the intensity and duration of cigarette smoking and HNSCC. The risk of cancer of the oral cavity for those who smoked between one and 15 cigarettes per day for one to 25 years was increased with an OR of 1.5 (95% CI: 1.4, 1.7). The OR for duration of 36 to 51 years and smoking one to 15 cigarettes per day was 3.7 (95% CI: 3.5, 3.9). An incremental effect was also reported by increasing both the intensity and the duration of smoking. The OR for those who smoked 26 to 40 cigarettes per day for 36 to 51 years was 8.4 (95% CI: 8.0, 8.9) (Di Credico et al. 2019). The increasing risk was reported for laryngeal cancer was even stronger. The OR of laryngeal cancer for those who smoked one to 15 cigarettes per day for 36 to 51 years was 8.9 (95% CI: 8.8, 9.1) and for those who smoked 26 to 40 cigarettes per day for 36 to 51 years was 33.9 (95% CI: 33.5, 34.3) (Di Credico et al. 2019).

Alcohol consumption

Hashibe et al. (2007) conducted a pooled analysis of individual-level data from 15 case-control studies including 1,024 cases and 15,227 controls to find out the association between alcohol consumption and the risk of HNSCC. All studies reported a dose-response association between the intensity of alcohol consumption and HNSCC. According to this pooled analysis study, the risk of pharyngeal and laryngeal cancers increased among those with highly frequent alcohol consumption (three or more drinks per day) compared to never-drinkers (OR: 2.0, 95% CI: 1.3, 3.2) (Hashibe et al. 2007).

Oral hygiene

Hashim et al. (2016) conducted a study to investigate the role of oral hygiene factors in the risk of HNSCC. In this study, they performed a pooled analysis using data from 13 studies participated in the INHANCE consortium, which included a total of 8,925 cases and 12,527 controls. The findings of the study revealed that the attributable fraction for oral hygiene in HNSCC was estimated to be 5.4% (95% CI: 0.4, 10%). This suggests that improvements in oral hygiene may have a modest protective effect against the risk of HNSCC. Different studies showed the positive association between different indices for oral hygiene and HNSCC such as no regular dental visits (OR: 2.9, 95% CI: 1.5, 5.6), brushing teeth less than two times per day (OR: 1.5, 95% CI: 1.0, 2.2), frequent gum bleeding (OR: 3.2, 95% CI: 1.4, 7.3), and tooth loss (OR: 2.3, 95% CI: 1.0, 5.1) (Chang et al. 2013; Divaris et al. 2010). Therefore, poor oral hygiene has been proposed as a risk factor for HNSCC, although the causality and independency of this factor are uncertain (Hashim et al. 2016)

Human papillomavirus (HPV)

Human papillomavirus (HPV) is causally associated with a varying percentage of cancers of the HNSCC (IARC 2006; Walboomers et al. 1999). The IARC has classified thirteen HPV types as carcinogens (de Sanjosé et al., 2018). The most common type of HPV responsible for head and neck cancers is HPV type 16 (de Sanjosé et al. 2018) which accounts for around 91% of oropharyngeal cancer and oral cavity cancer, and 53% of laryngeal cancer (Aupérin 2020; de Sanjosé et al. 2018). A recent systematic review of global

incidence trends in HNSCC showed that incidence trends increased for the HPV-related HNSCC subsites, regardless of age group, suggesting a consistent pattern across genders on a global scale (dos Santos Menezes et al. 2021). An epidemiological study showed that oral infection with high-risk HPV was significantly associated with HNSCC (OR: 2.4, 95% CI:1.1, 5.0) (Auguste et al. 2020). The prevalence of oral HPV infection in Iran is unknown.

Occupation

A recent INHANCE consortium publication on occupations and the risk of HNSCC using data from 12 case-control studies with 8,839 cases and 13,730 controls (Khetan et al. 2019) reported an increasing risk of HNSCC with a longer duration of employment for many occupations and an estimated population attributable fraction at 14.5% (95% CI: 7.1, 21.9%). The occupations with an increased risk of HNSCC included cooks (OR:1.4, 95% CI: 1.1, 1.7), waiters (OR:1.5, 95% CI: 1.2, 1.8), cleaners (OR:1.4, 95% CI: 1.1, 1.7), production and related workers including butchers and meat preparers (OR:1.6, 95% CI: 1.2, 2.1), occupations involving handling and production, spinners (OR:1.6, 95% CI: 1.1, 2.4), knitters (OR: 3.0, 95% CI: 1.4, 4.5), painters (OR:1.8, 95% CI: 1.4, 2.4), metal processors (OR:1.8, 95% CI: 1.2, 2.5), plumbers (OR:1.6, 95% CI: 1.3, 2.1), welders (OR:1.5, 95% CI: 1.2, 1.9), roofers (OR: 2.3, 95% CI: 1.4, 3.8), and material handling equipment operators (OR:1.8, 95% CI: 1.3, 2.4). These estimates were adjusted for age, sex, education level, race, region, alcohol intake and tobacco consumption (Khetan et al. 2019).

A study on occupations and the incidence of cancer in Nordic countries (Pukkala et al. 2009) showed that the standardized incidence ratios (SIR) for head and neck cancer (lip, oral cavity, tongue, salivary gland, pharynx, larynx) among men were the highest in waiters (SIR: 3.6, 95% CI: 3.3, 4.1), beverage workers (SIR: 2.2, 95% CI: 2.0, 2.7), cooks and stewards (SIR: 2.1, 95% CI: 1.8, 2.3), seamen (SIR: 1.7, 95% CI: 1.6, 1.8), economically inactive people (SIR: 1.5, 95% CI: 1.4, 1.5), and chimney sweeps (SIR: 1.4, 95% CI: 1.1, 1.9) (Table 2). Among men, the lowest SIRs (SIR<0.8) were observed in forestry workers, farmers, laboratory assistants, other health workers, launderers, religious workers, technical workers, physicians, teachers, and nurses.

The risk of head and neck cancer (lip, oral cavity, tongue, salivary gland, pharynx, larynx) among women was reported to be the highest in building hands (SIR:1.8, 95% CI: 1.5, 3.1), tobacco workers (SIR:1.7, 95% CI: 1.0, 2,4), beverage workers (SIR:1.6, 95% CI: 1.0, 2.5), journalists (SIR:1.6, 95% CI: 0.8, 2.0), and waiters (SIR:1.5, 95% CI: 1.3, 1.6) (Table 2). The lowest SIRs (SIR<0.8) of HNSCC among women were found in religious workers., other health workers, technical workers, nurses, physicians, farmers, shoe and leather workers, laboratory assistants, and forestry workers. Since the purpose of this study was to describe variation in cancer risks between occupational categories irrespective of whether this was related to direct occupational hazards or occupation typical life habites, the risk estimates are not adjusted for habits.

Table 2. Observed number (Obs), standardized incidence ratio (SIR) with 95% confidence intervals (CI) for head and neck cancer (lip, oral cavity, tongue, salivary gland, pharynx, larynx) in selected occupational categories in five Nordic countries, by sex. Recalculated from the results in https://astra.cancer.fi/NOCCA/.

Occupational category	Men		Women	
	Obs	SIR (95% CI)	Obs	SIR (95% CI)
Technical workers etc	3022	0.74 (0.71, 0.76)	70	0.83 (0.61, 1.01)
Laboratory assistants	54	0.81 (0.6, 1.04)	31	0.72 (0.44, 0.96)
Physicians	174	0.66 (0.55, 0.74)	18	0.79 (0.56, 1.5)
Dentists	88	0.85 (0.63, 0.98)	23	1.16 (0.93, 2.07)
Nurses	6	0.49 (0.15, 0.92)	261	0.80 (0.71, 0.92)
Artistic workers	361	1.30 (1.25, 1.53)	58	1.36 (1.06, 1.81)
Journalists	163	1.24 (1.07, 1.45)	29	1.58 (0.83, 1.97)
Administrators	2,434	0.89 (0.85, 0.93)	133	1.03 (0.87, 1.25)
Clerical workers	1,791	0.89 (0.85, 0.93)	2004	1.10 (1.07, 1.18)
Sales agents	2,498	1.04 (0.99, 1.07)	218	1.07 (0.90, 1.2)
Farmers	5,733	0.81 (0.79, 0.83)	423	0.77 (0.7, 0.86)
Fishermen	876	1.33 (1.27, 1.46)	6	1.61 (0.42, 3.22)
Forestry workers	998	0.84 (0.77, 0.88)	3	0.49 (0.07, 1.32)
Drivers	3,396	1.18 (1.17, 1.25)	42	1.21 (0.69, 1.4)
Shoe and leather workers	220	1.07 (0.93, 1.22)	37	0.75 (0.67, 1.26)
Building hands	2,381	1.26 (1.19, 1.30)	27	1.83 (1.45, 3.13)
Printers	495	1.05 (0.94, 1.13)	71	1.26 (0.94, 1.57)
Chemical process workers	757	1.07 (1.01, 1.16)	41	0.87 (0.75, 1.37)
Food workers	1,028	1.09 (1.03, 1.17)	244	1.1 (0.96, 1.26)
Beverage workers	145	2.23 (1.96, 2.72)	20	1.59 (1.0, 2.54)
Tobacco workers	14	1.20 (0.59, 1.86)	23	1.65 (0.98, 2.43)
Waiters	336	3.58 (3.27, 4.06)	351	1.53 (1.29, 1.62)
Building caretakers	713	1.12 (1.03, 1.19)	1210	1.15 (1.11, 1.25)

Hormonal factors in women

An INHANCE study based on 11 studies from Europe, North America, and Japan pooled the data of 1,572 female cases and 4,343 controls to find out the association between hormonal factors and the risk of HNSCC among females. Based on this pooled analysis, the study suggested an inverse association between female hormones and the risk of HNSCC (Hashim et al. 2017).

After adjusting for tobacco smoking and alcohol drinking, a decreased risk was reported for the women who used hormone replacement therapy (OR = 0.58; 95% CI: 0.34, 0.77). In addition, pregnancy and giving birth at an age below 35 years old showed a protective effect on HNSCC risk with ORs of 0.6 (95% CI: 0.4, 0.9) and 0.6 (95% CI: 0.4, 0.9), respectively (Hashim et al. 2017).

Physical activity

Studies about the association between physical activity and HNSCC have reported a protective effect of physical activity on head and neck cancer risk. A study using 246 cases and 504 controls in the United States reported a positive association between recreational physical inactivity and HNSCC (OR: 2.7, 95% CI: 1.9, 4.0) and suggested that lifelong physical inactivity is associated with HNSCC (Platek et al. 2017).

Another study conducted by Lin et al. in Taiwan using 623 cases and 731 controls found that the association of HNSCC risk and recreational physical activity decreased by 30% (OR: 0.7, 95% CI: 0.5, 0.8) (Lin et al. 2017). A prospective cohort study discovered that three or more hours of weekly physical activity was linked to a 40% reduction in the risk of HNSCC (OR: 0.6, 95% CI: 0.4, 1.0) (Hashibe et al. 2013). Another study the analysed data from the INHANCE consortium on 2,289 cases and 5,580 controls to investigate the association between recreational physical activity and head and neck cancer suggested that engaging in moderate physical activity can decrease the risk of head and neck cancer by 20% when compared to those who reported no or very low physical activity (OR: 0.8, 95% CI: 0.7, 0.9). Additionally, this study observed a lower risk of oral cavity cancer (OR: 0.7, 95% CI: 0.6, 1.0) and pharyngeal cancer (0.7, 95% CI: 0.5, 1.0) among individuals who engaged in moderate physical activity compared to those who reported low physical activity (Nicolotti et al. 2011).

Nutrition

Epidemiological studies have been conducted to evaluate the effect of diet on the risk of HNSCC (Aupérin 2020). An inverse association of consuming fruit and vegetables on HNSCC has been reported in many studies (Chuang et al., 2012; Edefonti et al., 2010 a-b; Lagiou et al., 2009). One large sample size study conducted by Chuang et al. using INHANCE data investigated the association between diet and HNSCC (Chuang et al. 2012). This study included pooling the data of 22 case-control studies with 14,520 cases and 22,737 controls and showed an inverse association between a higher frequency of fruit intake (OR:0.5, 95% CI: 0.4, 0.6) and vegetable intake (OR: 0.7, 95% CI: 0.5, 0.9) on the risk of HNSCC (Chuang et al. 2012). On the other hand, several studies suggested meat intake as a risk factor for HNSCC (Edefonti, et al., 2010 a-b), but this association was not consistent (Peters et al. 2008; Sapkota et al. 2008). The INHANCE study (Chuang et al. 2012) showed a positive association between red meat and processed meat consumption and HNSCC risk (OR: 1.4, 95% CI: 1.1, 1.7) and (OR:1.4, 95% CI: 1.1, 1.7), respectively.

Socioeconomic status

Low socioeconomic status (SES), often characterized by low education or income, is associated with a higher risk of HNSCC (Conway et al. 2015; Stanford-Moore et al. 2018). The large INHANCE study investigated the effect of education and income as a proxy of SES on the risk of HNSCC (Conway et al. 2015). The study pooled the data of 31 case-control studies from 27 countries including 23,964 cases and 31,954 controls and showed that low education was associated with an increased risk of HNSCC (OR: 2.5, 95% CI: 2.0, 3.1) (Conway et al. 2015). A survey conducted among working-age Finns (Pukkala, 1995), which included 109,000 cancer cases, revealed that men belonging to the lowest social class (out of four) showed an almost five-fold incidence if lip cancer as compared to the highest class. Among women, the highest incidence of lip cancer was also observed in social class IV, with an SIR of 2.2 (95% CI: 1.5, 3.1) as compared to the population average. In tongue cancer, there was no noticeable social class variation. The incidence of oral cavity cancer in women exhibited a clear upward trend with increasing social status. The SIR for social class I was 1.6 (0.9, 2.7), whereas, for social class IV, it was 0.7 (0.4, 1.1). Among men there was no regular trend in incidence of oral cavity cancer by social class. The study considered nasopharyngeal cancer separately from other types of pharyngeal cancer due to differences in their causes. The incidence of nasopharyngeal cancer among men in the highest social class was more than three times higher compared to the lowest class (Pukkala, 1995).

1.2 Bladder cancer

The bladder is a hollow organ to store urine. The bladder contains several layers that, together, constitute the bladder wall. The innermost layer of the bladder is called the urothelial or mucosal layer (Pashos et al. 2002). Bladder cancer (BC) generally originates from the urothelial layer which is the most common (75%) type of BC (Lenis, Lec, and Chamie 2020; Pashos et al. 2002; Sanli et al. 2017).

Generally, urothelial carcinoma includes tumors of the bladder, upper urinary tract (renal pelvis and ureters), and proximal urethra. Approximately 90% to 95% of urothelial carcinoma is located in the bladder (Lenis, Lec, and Chamie 2020). The ICD-O-3 code for the BC is C67.

1.2.1 Epidemiology of bladder cancer

1.2.1.1 Occurrence

BC is the 10th most common cancer worldwide with 57,3000 new cases and 21,3000 deaths each year (Sung et al. 2021). BC is about four times higher among men than women. The ASR and ASMR for men are 9.5 and 3.3 per 100,000 person-years worldwide (Figures 5 and 6).

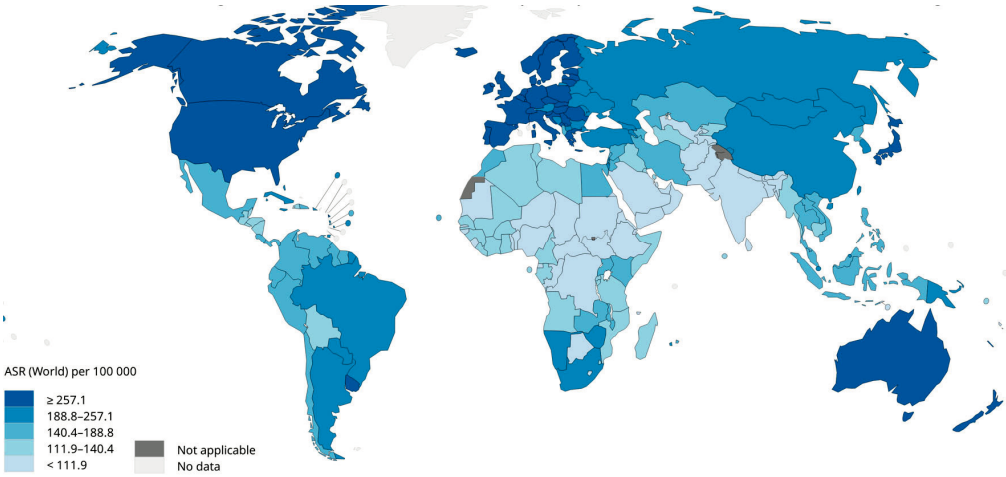

Figure 5. Estimated incidence rates (ASR) of bladder cancer, by country, age-standardized to the world standard population (GLOBOCAN-2020) ("IARC" 2020).

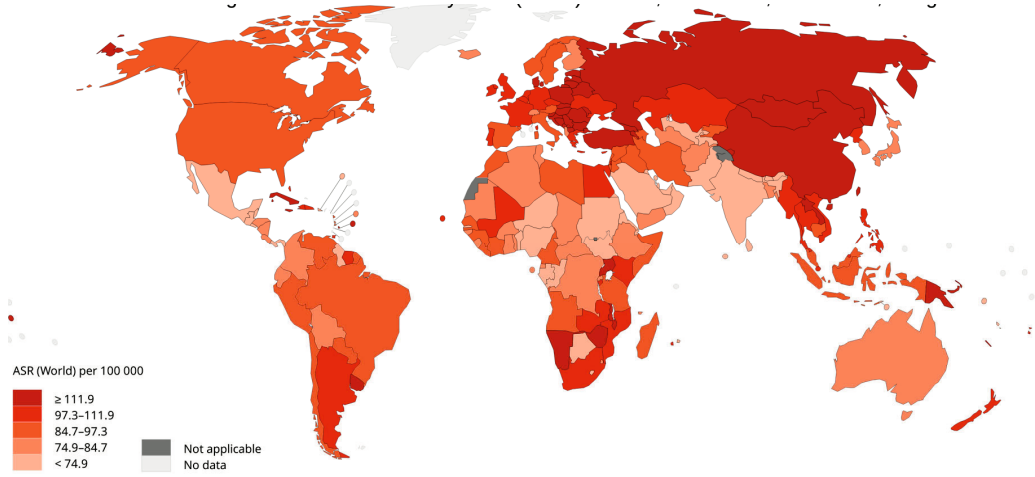

Figure 6. Estimated mortality rates (ASR) of bladder cancer, by country, age-standardized to the world standard population (GLOBOCAN-2020) ("IARC" 2020).

BC is the sixth most common cancer in Iran with 5,065 new cases and 1,760 deaths annually ("IARC" 2020; Kalan Farmanfarma, Mahdavifar, and Salehiniya 2020). In addition, BC is the fourth most common cancer among men in Iran with 4,282 new cases and an age-standardized (World Standard) ASR of 14.3/100 000 in 2014 (Ferlay et al. 2023; Kalan Farmanfarma, Mahdavifar, and Salehiniya 2020; Roshandel et al. 2019).

Figure 7 presented the ASRs of BC per 100,000 person-years, stratified by sex, over five years in Iran and the ASRs of BC from 1977 to 2020 in the Nordic countries. The ASR increased from 2.1 to 3.8 per 100,000 person-years in women and from 8.4 to 14.4 per 100,000 person-years in men from 2003 to 2008 in Iran. The annual change in percentage of the ASR was 11.5 (95% CI: 9.0, 14.0) in women and 10.8 (95% CI: 8.0, 13.6) in men, an increase which might be attributed to population growth, smoking, and shifts in lifestyle (Rafiemanesh et al. 2018). According to the NORDCAN statistics, the ASR of BC in men fluctuated from 1977 to 2015 while subsequently decreasing from 2016 to 2020. The ASR of BC in women does not show a big change over three decades and decreased from 2018 to 2020 (Figure 7).

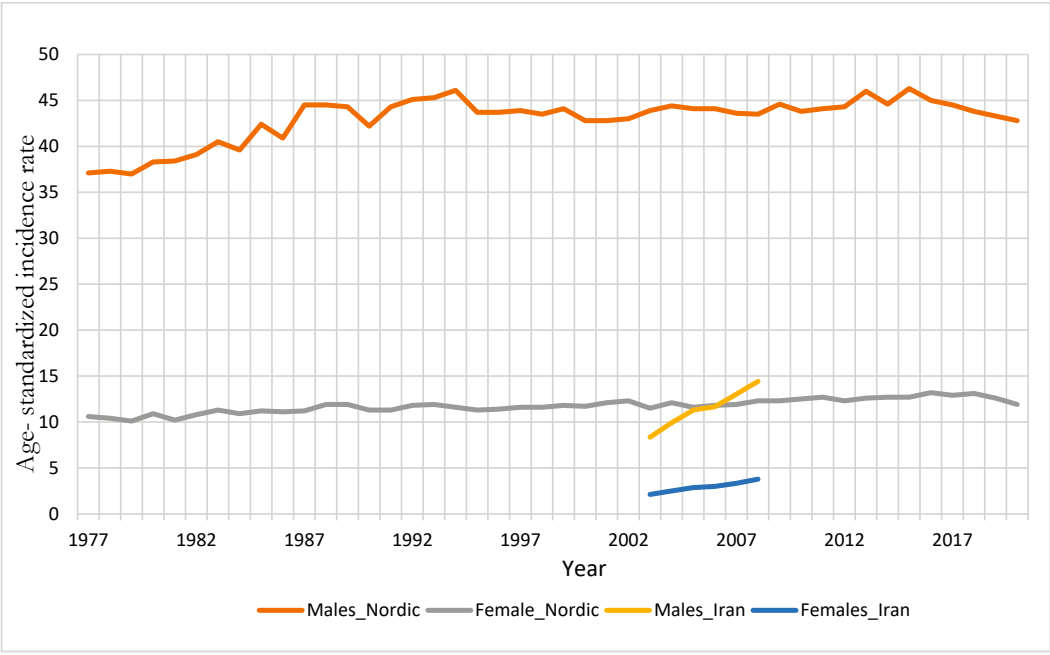

Figure 7. Age-standardized incidence rates (per 100,000 persons-years of bladder cancer in the Golestan province in Iran (Rafiemanesh et al. 2018) and in the Nordic countries in the period 1977–2020 (*https://Nordcan.Iarc.Fr/En*, 2023), by sex.

1.2.1.2 Etiological factors

Different epidemiological studies carried out over decades of research have shown different identified risk factors for BC including sex, age, occupation, alcohol drinking, nutrition, and tobacco smoking (Lenis, Lec, and Chamie 2020; Pashos et al. 2002; Saginala et al. 2020).

Sex

BC is approximately four times more common among men than women. Likewise, mortality due to BC is four times greater in men than women (Sung et al. 2021). The sex difference might be due to different rates of risk factors among men like tobacco smoking (World Health Organization, 2012; Ramström & Wikmans, 2014). The occurrence of BC is also high among women in countries with a high rate of tobacco use among women, such as Lebanon, which has the highest rate of BC among women globally. (Saginala et al. 2020). With the increasing prevalence of smoking among women, around 39% of BC cases among women were attributed to smoking in 2014 in the United States in comparison with 49% of new cases attributed to smoking among men (Sung et al. 2021).

Age

BC is predominantly an adult disease, with the majority of diagnosed cases found in those over 55 years old; almost 80% of BC diagnoses in the United States are in patients over 65 years old with an average age

of onset at 73 years old (Saginala et al. 2020; Siegel, Miller, and Jemal 2019). BC rarely occurs in children and young adults, but when it does occur, it is often noninvasive and low-grade. Thus, older age at the diagnosis of BC may be related to a prolonged exposure required for the carcinogenic effects of various mutagens to take effect (Linn et al. 1998).

Tobacco smoking

Tobacco smoking is one of the most important risk factors for BC, accounting for around 50% to 65% of new BC cases annually (Saginala et al. 2020). Different epidemiological studies have shown that smoking increases the risk of BC by three to four times (Al Hussein Al Awamlh et al. 2019; Freedman et al. 2011; Saginala et al. 2020). The pooled results of 15 case-control studies showed that current smokers had a much higher risk of BC risk compared to never-smokers (OR: 2.2, 95% CI: 2.1, 2.4) (van Osch et al. 2019). A national longitudinal study from the USA showed that 65% of those who died due to BC were ever-smokers and the hazard ratio (HR) of those who had smoked at any point in their lives was 1.9 (95% CI: 1.3, 3.0) (Al Hussein Al Awamlh et al. 2019).

Environmental and occupational exposure

One of the most important preventable risk factors for BC is occupational exposure. The risk of BC is increased by different industrial chemicals including chlorinated hydrocarbons, aromatic amines, polycyclic aromatic hydrocarbons, chromates, dinitrotoluene, arsenic, beryllium, cadmium, nickel, wood dust, crystalline silica, brown coal phosphors, furnace emissions, smoke from diesel engine, ionizing radiation and non-ionizing radiation, thermal shock, asbestos, pesticides, aniline, and aniline-based or benzidine-based dyes (Deb et al. 2019; IARC 2006; Pedroso et al. 2022; Saginala et al. 2020). A recent systematic review and meta-analysis about occupational exposure and the risk of BC showed a significant effect of this exposure on the risk of BC (pooled OR: 1.5, 95% CI: 1.4, 1.8) (Deb et al. 2019). Another pooled analysis using two Italian case-control studies reported a significantly increased risk of BC for chemical engineering technicians, postmen, and lathe operators (Sciannameo et al. 2019). The analysis also reported that ever exposure to cadmium increased the risk of BC compared to never exposure (OR: 1.3, 95% CI: 1.0, 1.6) (Sciannameo et al. 2019). In a study about occupation and cancer in five Nordic countries, the highest SIR among men for BC between occupation categories was for waiters (SIR: 1.5, 95% CI: 1.3, 1.7), chimney sweeps (SIR: 1.5, 95% CI: 1.2, 1.8), hairdressers (SIR: 1.3, 95% CI: 1.2, 1.5), assistant nurses (SIR: 1.3, 95% CI: 1.1, 1.5), and seamen (SIR: 1.2, 95% CI: 1.2, 1.3). The lowest SIRs (SIR≤0.80) among men were reported for occupation categories including teachers, gardeners, forestry workers, nurses, domestic assistants, and farmers. Similarly, the highest SIR for women was reported for tobacco workers (SIR: 2.0, 95% CI: 1.5, 2.7), waiters (SIR: 1.4, 95% CI: 1.3, 1.6), and chemical process workers (SIR: 1.3, 95% CI: 1.1, 1.6), and the lowest SIR was observed among engine operators, smelting workers, welders, woodworkers, gardeners, physicians, dentists, farmers, and journalists (Table 3) (Pukkala et al. 2009).

Table 3. Observed number (Obs), standardized incidence ratio (SIR) with 95% confidence intervals (95% CI) for bladder cancer (BC) in selected occupational categories in five Nordic countries, by sex.

Occupational category	Men		Women	
	Obs	SIR (95% CI)	Obs	SIR (95% CI)
Technical workers. etc	7679	1.02 (1.00, 1.05)	126	1.08 (0.90, 1.28)
Laboratory assistants	124	1.11 (0.93, 1.32)	58	1.05 (0.79, 1.35)
Physicians	505	1.03 (0.94, 1.12)	24	0.76 (0.49, 1.13)
Dentists	214	1.10 (0.96, 1.26)	21	0.71 (0.44, 1.08)
Artistic workers	594	1.17 (1.08, 1.27)	72	1.13 (0.88, 1.42)
Journalists	238	1.02 (0.89, 1.15)	15	0.61 (0.34, 1.01)
Administrators	5622	1.07 (1.04, 1.10)	251	1.20 (1.06, 1.36)
Sales agents	5095	1.16 (1.12, 1.19)	405	1.26 (1.15, 1.39)
Farmers	9447	0.68 (0.67, 0.70)	652	0.66 (0.62, 0.72)
Gardeners	2491	0.77 (0.74, 0.81)	671	0.76 (0.71, 0.82)
Fishermen	1450	1.14 (1.08, 1.20)	3	0.48 (0.10, 1.40)
Forestry workers	1599	0.74 (0.71, 0.78)	6	0.59 (0.22, 1.28)
Miners and quarry workers	480	0.94 (0.86, 1.03)	3	1.61 (0.33, 4.72)
Transport workers	2107	1.08 (1.03, 1.12)	31	1.24 (0.84, 1.75)
Drivers	5973	1.15 (1.12, 1.18)	64	1.20 (0.93, 1.54)
Postal workers	1106	1.07 (1.01, 1.14)	439	1.06 (0.97, 1.17)
Welders	822	1.06 (0.99, 1.13)	4	0.80 (0.22, 2.04)
Woodworkers	5698	0.94 (0.91, 0.96)	56	0.79 (0.60, 1.02)
Painters	1642	1.08 (1.03, 1.14)	15	1.52 (0.85, 2.51)
Building hands	3592	1.02 (0.98, 1.05)	15	0.87 (0.49, 1.44)
Bricklayers	976	1.03 (0.97, 1.10)	2	3.16 (0.38, 11.41)
Printers	1018	1.19 (1.12, 1.27)	126	1.46 (1.22, 1.74)
Chemical process workers	1389	1.05 (1.00, 1.11)	103	1.29 (1.07, 1.57)
Tobacco workers	29	1.20 (0.80, 1.72)	50	2.01 (1.49, 2.65)
Waiters	249	1.50 (1.32, 1.69)	554	1.44 (1.32, 1.56)
Building caretakers	1276	1.10 (1.04, 1.16)	1946	1.09 (1.04, 1.14)

Alcohol consumption

Several epidemiological studies have evaluated the effect of alcohol consumption on BC risk but the risk of alcohol consumption has not been proven to be strong (Larsson et al. 2020). A systematic review and meta-analysis (Pelucchi et al. 2012) evaluated the risk of BC at different levels of alcohol consumption and concluded that the relative risk (RR) of BC for moderate alcohol drinkers was 1.0 (95% CI: 0.9, 1.1) and for heavy drinkers 1.0 (95% CI: 0.8, 1.3) compared to non-drinkers.

Red meat

Some studies reported that red and processed meat seems to increase the risk of BC (Aveta et al. 2022; Wang and Jiang 2012). A recent review related to the impact of meat intake on BC highlighted a positive association between BC and red meat and processed meat intake, particularly salami, pastrami, corned beef and bacon (Aveta et al. 2022). Another study used the pooled data from 11 cohort studies including 2,848

BC cases and 515,697 controls with a total of 5,498,025 person-years of follow-up to evaluate the association between meat and fish consumption on BC and found an increased risk of BC for high intake (highest tertile) organ meat compared to low intake (lowest tertile) with an HR of 1.2 (95% CI: 1.0, 1.4, p-trend=0.03) (Dianatinasab et al. 2021). Meanwhile, a large European investigation into cancer and nutrition (EPIC) showed that there is no overall association between red meat intake and the risk of BC (HR:1.1, 95% CI: 1.0, 1.1) (Jakszyn et al. 2011).

1.3 Colorectal cancer

The colorectal system includes the colon and rectum segments of the intestine. The colon is one of the main parts of the digestive tract and is the first and largest part of the large intestine. This segment is located between the small intestine and the rectum. The colon's principal function is the absorption of nutrients, minerals, and water and it is used to pass waste materials. This segment consists of four sections, which include the ascending colon and the transverse colon on the right side; and the descending colon and the sigmoid colon ending with the rectum on the left side (Figure 8) (Dekker et al., 2019; *U. S. National Institutes of Health, National Cancer Institute*, 2023).

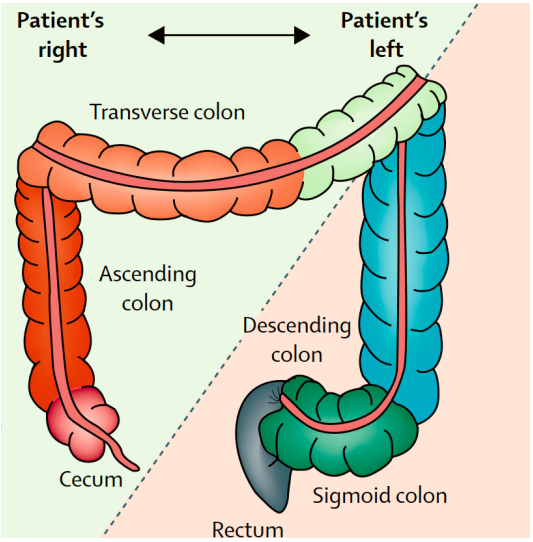

Figure 8. Anatomical subtypes of colorectal cancer (Dekker et al. 2019).

Colorectal cancer (CRC) usually grows slowly and generally does not show any symptoms until the tumor becomes so big that it blocks the passage of feces, and leads to cramping, pain, or bleeding that can present as visible bleeding with bowel movements (Simon 2016). The ICD-O-3 codes for the colon are C18.0-C18.9, rectosigmoid junction C19.9 and rectum C20.9.

1.3.1 Epidemiology of colorectal cancer

1.3.1.1 Occurrence

CRC is the 10th most common cancer worldwide with 573,000 new cases each year and 213,000, annual deaths (Sung et al. 2021). CRC is more than 4 times higher among men than women. The ASR and ASMR for men are 9.5 and 3.3 per 100,000 person-year worldwide (Figure 9 and 10) ("IARC" 2020; Sung et al. 2021).

CRC is the third most common cancer in Iranian men with 5,644 new cases each year and an ASR of 16.6 per 100,000 person-year (Roshandel et al. 2019). In addition, CRC is the second most common cancer among women in Iran with 4,217 new cases annually and an ASR of 11.9 per 100,000 person-years (Roshandel et al. 2019).

According to the data from the Golestan province, the incidence of CRC increased among both men and women from 2004 to 2013 (Figure 11). In Nordic countries the incidence of CRC has been decreasing over the past decades among both men and women (Figure 11).

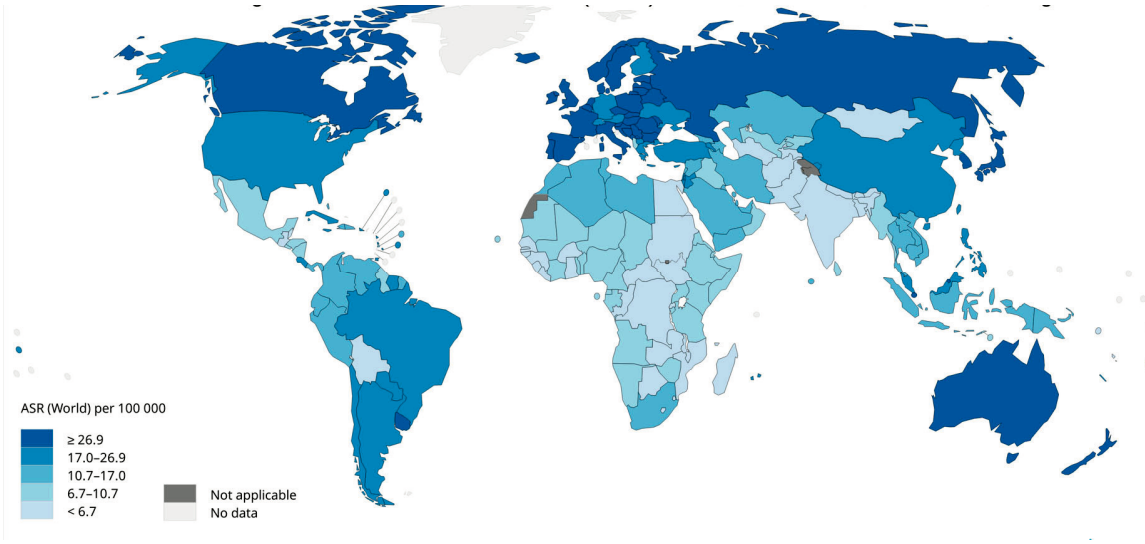

Figure 9. Estimated incidence rates (ASR) of colorectal cancer, by country, age-standardized to the world standard population (GLOBOCAN- 2020) ("IARC" 2020).

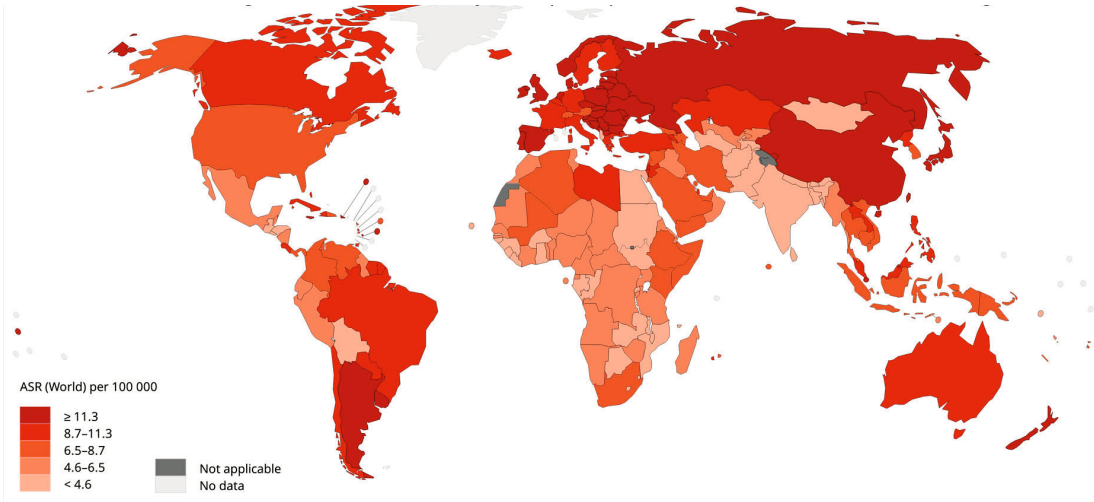

Figure 10. Estimated mortality rates (ASR) of colorectal cancer, by country, age-standardized to the world standard population (GLOBOCAN- 2020) ("IARC" 2020).

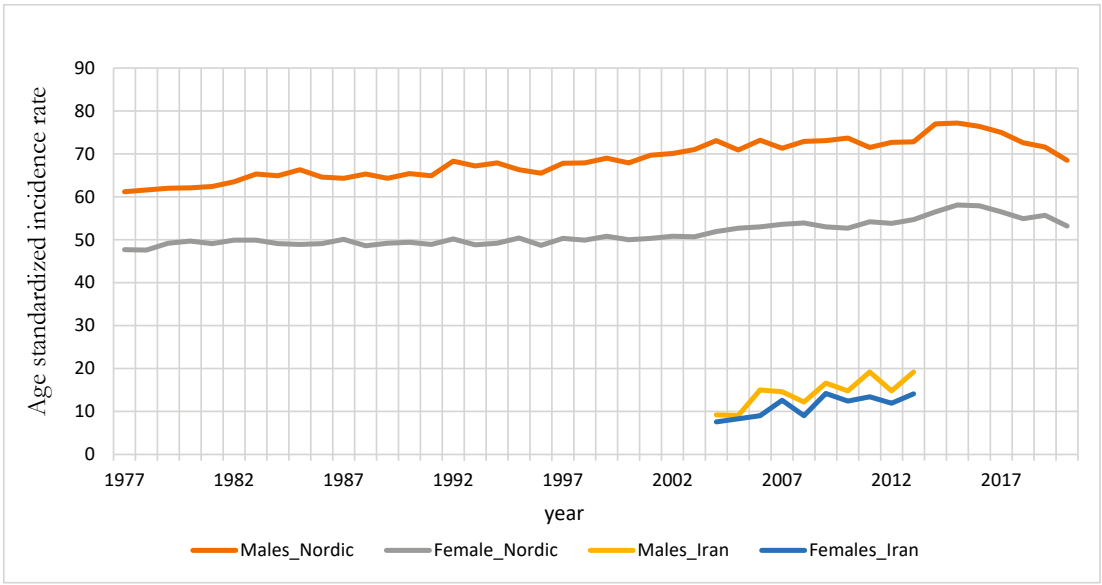

Figure 11. Age-standardized incidence rates (per 100,000 persons-years of colorectal cancer in the Golestan province of Iran (Hasanpour-Heidari et al. 2019) and in the Nordic countries in the period 1977–2020 (*https://Nordcan.Iarc.Fr/En*, 2023), by sex.

1.3.1.2 Etiological factors

Sex

According to the American Cancer Society report, the incidence of CRC among men is higher than among women ("American Cancer Society" 2020). The incidence among men is 60% higher than among women for rectal cancer and 20% higher than among women for colon cancer ("American Cancer Society" 2020). This disparity is probably due to differences in exposure to different risk factors, for instance, cigarette smoking, drinking alcohol and sex hormones (Murphy et al. 2011). However, there is no gender difference in the incidence of CRC among persons younger than 45 years old ("American Cancer Society" 2020). However, women are more susceptible to right-sided colon cancer than men, which is more aggressive than left-sided colon cancer (Rawla, Sunkara, and Barsouk 2019; Kim et al. 2015).

Age

Age is a well-established risk factor for CRC worldwide. The majority of cases are diagnosed in individuals over the age of 50, with the incidence increasing sharply after the age of 60 ("American Cancer Society" 2020; Sawicki et al. 2021). Individuals who are 65 years and above have an estimated three times higher risk of developing CRC compared to those aged 50–64, and a 30 times higher risk than those aged 25–44 (Rawla, Sunkara, and Barsouk 2019). CRC is particularly linked to age-related factors, especially in developed countries where the incidence rate is the highest.

Family history

A family history of CRC in first-degree relatives significantly increases the risk of developing this cancer (Sawicki et al. 2021). Up to 30% of CRC patients reported a family history of CRC which means there are predisposing germline mutations which have not been identified yet. Those patients with a positive family history of CRC in their first-degree relatives were at a two to four times higher risk (De Rosa et al. 2015; Slattery et al. 2003). A higher risk of CRC has been reported for those with relatives diagnosed before the age of 50 years and for those with more than one family member with CRC (Slattery et al. 2003; Kerber et al. 1998; Slattery and Kerber 1994).

A positive family history of cancer for CRC is defined as having relatives with a history of cancer in cancer types that are possibly linked to Lynch syndrome or hereditary nonpolyposis colorectal cancer (HNPCC). HNPCC is an autosomal dominant genetic condition that is associated with a high risk of colon cancer and cancers of the endometrium (second most common), ovaries, stomach, small intestine, hepatobiliary tract, upper urinary tract, brain, and skin (Peltomäki, Olkinuora, and Nieminen 2020; Hampel et al. 2005).

Inflammatory bowel disease

Inflammatory bowel disease (IBD) refers to those with Crohn's disease and ulcerative colitis which are chronic relapsing disorders of unknown origin and varying severity (Jess et al. 2007). Patients with a history of chronic IBD have a two times higher risk of developing CRC (Rawla, Sunkara, and Barsouk 2019).

However, some studies reported that the risk of CRC declined for those with IBD (Jess et al. 2007; Kewenter, Ahlman, and Hulten 1978). The recent systematic review related to the risk of CRC in IBD showed that the pooled SIR of CRC in all IBD patients was 1.7 (95% CI: 1.2, 2.2), and the SIR for the high-risk group with extensive colitis and IBD before age 30 was 6.4 (95% CI: 2.4, 17.5), and 7.2 (95% CI: 2.9, 17.8), respectively (Lutgens et al. 2013).

Overweight and obesity

The incidence of both obesity and CRC has been increasing in recent years (Elangovan et al. 2021; Ye et al. 2020). Body mass index (BMI) is used as a standard value derived from the person's weight (in kilograms) divided by the body height (in meters square) in both genders. A person is considered overweight with a BMI ≥ 25 and obese with a BMI ≥ 30 (Ye et al. 2020). A study about the association of obesity with the risk of early-onset CRC among women showed that the risk of CRC increased with weight gain: the RR of early-onset CRC was 1.6 for those with a BMI of 23 or greater (RR: 1.6, 95% CI: 1.0, 2.6) compared with women with a BMI of 18.5 to 20.9 (Liu et al. 2019). Another study showed that the risk of CRC was 40% higher for those with a BMI ≥ 30 compared to those with a BMI < 30 in the age group between 18 to 49 years old (OR: 1.4, 95% CI: 1.0, 1.9), and they reported that there is significant interaction between age and BMI (P-value=0.02) (Sanford et al. 2020).

Physical activity

Lack of physical activity has been associated with a higher risk of CRC in epidemiological studies; however, it is not clear whether this association is confounding or causal (Papadimitriou et al. 2020). The recent review showed that those who are regularly physically active have a 25% lower risk of CRC. Meanwhile, those with a lack of physical activity (sedentary) have up to 50% higher risk of developing CRC (Rawla, Sunkara, and Barsouk 2019). Another study showed that the highest level of physical activity was inversely associated with the risk of CRC compared to the lowest level (Robsahm et al. 2013); the RR of both proximal and distal colon cancer was 0.8. A study on the perceived physical workload (PPWL) indicated a stronger protective impact on colon cancer in the highest PPWL category for men (OR: 0.7, 95% CI: 0.7, 0.8) than for women (OR: 0.9, 95% CI: 0.7, 1.0) as compared to lowest PPWL category (Sormunen et al. 2016). No association was found between physical activity and the risk of rectal cancer (RR: 1.0, 95% CI: 0.9, 1.1) in Norway (Robsahm et al. 2013). There was a slightly protective effect against rectal cancer among Nordic men (OR: 0.87, 95% CI: 0.85, 0.90) and women with a high PPWL (OR: 0.93, 95% CI: 0.83, 1.04) compared to those with a low PPWL (Sormunen et al. 2016).

Red meat and processed meat

According to the IARC monograph, red meat and processed meat were considered as probably carcinogenic to humans (Group 2A) and carcinogenic to humans (Group 1), respectively (IARC 2015). Different epidemiological studies have shown that there is convincing evidence to support a positive association between the intake of red and processed meat and the risk of CRC (Farvid et al. 2021; Kossenas and Constantinou 2021; Mattiuzzi and Lippi 2021; Mehta et al. 2020). A recent systematic review of red and processed meat intake and the risk of cancer showed that red meat intake was associated with an increased risk of CRC (RR: 1.1, 95% CI: 1.0, 1.2), colon cancer (RR: 1.2, 95% CI: 1.1, 1.3) and rectal cancer

(RR: 1.2, 95% CI: 1.0, 1.5). Moreover, processed meat consumption was associated with an 18% higher risk of CRC, a 21% higher risk of colon cancer and a 22% higher risk of rectal cancer (Farvid et al. 2021). Another recent systematic review on the association between diet and cancer from the EPIC study reported that a higher intake of red and processed meat increased the risk of CRC (Ubago-Guisado et al. 2021).

Smoking

Tobacco smoking is identified as an established risk factor for the development of different types of cancer including CRC (Botteri et al. 2020; Rawla, Sunkara, and Barsouk 2019). A recent meta-analysis showed that a pooled RR for the risk of CRC for current smokers was 1.4 (95% CI: 1.1, 1.2) and for former smokers was 1.2 (95% CI: 1.1, 1.2) compared to never smokers (Botteri et al. 2020). It was also shown that CRC risk increased linearly with smoking intensity. Those who had stopped smoking for more than 25 years had a significantly reduced risk of CRC in comparison with current smokers (Botteri et al. 2020). Another study to evaluate the associations of smoking with early and late onset CRC showed a strong association for both early and late onset CRC. The risk of early onset CRC for current smokers was 1.6 (95% CI: 1.2, 2.0), and for former smokers was 1.4 (95% CI: 1.1, 1.8). Similarly, the risk of late-onset CRC for current smoking was 1.5 (95% CI: 1.3, 1.7), and for former smokers was 1.2 (95% CI: 1.1, 1.4) (Li et al. 2023).

Alcohol

Alcohol consumption is considered one of the established risk factors for CRC and it is estimated that drinking alcoholic beverages between two and three times per day increases the risk of CRC by about 20%, and drinking more than three times per day this risk increases it by about 40% (Sawicki et al. 2021). Those who used to drink alcoholic beverages four times or more per day risk of developing CRC increases up to 52% (Marley and Nan 2016). The EPIC study in which 478,732 free cancer subjects were followed up for 6.2 years, during which 1,833 CRC occurred showed that lifetime alcohol intake increased the risk of CRC (HR: 1.1, 95% CI:1.0, 1.1), and showed an increased risk of rectal cancer (HR: 1.1, 95% CI: 1.06, 1.18) (Ferrari et al. 2007). Also, a meta-analysis of 16 studies of the association between alcohol intake and CRC showed that very heavy alcohol consumption (more than three drinks per day) was associated with an increased risk of CRC (OR: 1.3, 95% CI: 1.1, 1.4) (McNabb et al. 2020).

Occupation

Occupational exposure is not considered an etiological factor for CRC; however, a greater risk of CRC has been reported in several occupational groups such as employees working in the textile industry (Mastrangelo et al. 2002), automobile industry (Swanson, Belle, and Burrows Jr 1985) and beverage industry (Garabrant et al. 1984), as well as the individuals exposed to asbestos (Baan et al. 2009), wood dust (Dement et al. 2003), organic solvents (Dumas et al. 2000), dioxin (Bertazzi et al. 2001), and metalworking fluids (Calvert et al. 1998). Another study showed that the risk of CRC increased for workers in industries involving leather (RR: 1.7, 95% CI: 1.2, 2.3), basic metals (RR: 1.3, 95% CI: 1.1, 1.7), plastic manufacturing (RR: 1.3, 95% CI: 1.0, 1.7), rubber manufacturing (RR: 1.3, 95% CI: 0.9, 1.8), and exposure to asbestos (RR: 1.4, 95% CI: 1.1, 1.8) (Oddone, Modonesi, and Gatta 2014).

A study concerning the relation between occupation and cancer in Nordic countries reported that the incidence of colon cancer was the lowest in occupational categories and it is mostly related to sedentary work (Pukkala et al. 2009). According to this study, the highest SIR for CRC for men was reported among waitresses (SIR: 1.4, 95% CI: 1.2, 1.5), chimney sweeps (SIR: 1.3, 95% CI: 1.1, 1.6), and beverage workers (SIR: 1.3, 95% CI: 1.2, 1.5). The lowest SIR was reported for gardeners, farmers, and forestry workers. Moreover, the highest SIR for the CRC among women was reported for chimney sweeps (SIR: 3.0, 95% CI: 1.3, 9.7), tobacco workers (SIR: 1.2, 95% CI: 1.0, 1.4), and printers (SIR: 1.1, 95% CI; 1.1, 1.3), and the lowest SIR for occupations such as engine operators and smelting workers (Table 4) (Pukkala et al. 2009).

Table 4. Observed number (Obs), standardized incidence ratio (SIR) with 95% confidence intervals (95% CI) for colorectal cancer (CRC) in selected occupational categories in five Nordic countries, by sex.

Occupational category	Men		Women	
	Obs	SIR (95% CI)	Obs	SIR (95% CI)
Laboratory assistants	195	1.05 (0.92, 1.22)	243	0.89 (0.78, 1.01)
Physicians	821	1.03 (0.96, 1.10)	162	1.04 (0.89, 1.22)
Dentists	351	1.08 (0.97, 1.20)	163	1.12 (0.96, 1.31)
Forestry workers	2955	0.8 (0.77, 0.83)	46	0.87 (0.66, 1.02)
Miners and quarry workers	800	0.91 (0.85, 0.97)	2	0.21 (0.04, 0.73)
Seamen	2282	1.10 (1.05, 1.14)	3	0.65 (0.19, 2.04)
Transport workers	3292	1.04 (1.01, 1.08)	144	1.11 (0.94, 1.31)
Drivers	9184	1.08 (1.06, 1.11)	245	0.96 (0.85, 1.09)
Postal workers	1765	1.05 (1.00, 1.10)	2185	1.04 (1, 1.09)
Textile workers	1911	1.08 (1.03, 1.13)	5137	1.07 (1.04, 1.1)
Shoe and leather workers	756	1.13 (1.05, 1.21)	387	0.98 (0.89, 1.08)
Smelting workers	2914	1.02 (0.98, 1.06)	65	0.79 (0.62, 1)
Plumbers	1566	1.11 (1.05, 1.16)	2	0.74 (0.17, 3.3)
Bricklayers	1546	1.02 (0.97, 1.08)	2	0.58 (0.11, 2.2)
Printers	1599	1.14 (1.08, 1.19)	491	1.14 (1.05, 1.25)
Chemical process workers	2237	1.00 (0.96, 1.05)	425	1.11 (1.01, 1.23)
Beverage workers	254	1.31 (1.16, 1.49)	85	0.86 (0.7, 1.07)
Tobacco workers	41	1.10 (0.81, 1.49)	129	1.16 (0.97, 1.38)
Glassmakers etc.	2299	0.99 (0.95, 1.04)	910	1.06 (0.99, 1.13)
Packers	4606	1.09 (1.06, 1.13)	1419	1.04 (1.04, 1.15)
Engine operators	3635	1.04 (1.01, 1.07)	74	0.83 (0.66, 1.04)
Public safety workers	2690	1.15 (1.11, 1.19)	121	1.1? (0.93, 1.32)
Cooks and stewards	464	1.04 (0.95, 1.14)	1911	1.02 (0.97, 1.06)
Domestic assistants	19	1.12 (0.74, 1.82)	4696	0.96 (0.94, 0.99)
Waiters	379	1.37 (1.24, 1.52)	2107	1.08 (1.03, 1.12)
Building caretakers	1974	1.05 (1.01, 1.10)	8372	0.96 (0.94, 0.98)
Chimney sweeps	155	1.32 (1.13, 1.55)	4	3.0? (1.27, 9.65)
Hairdressers	474	1.09 (0.99, 1.19)	806	1.06 (0.99, 1.13)
Launderers	265	0.97 (0.87, 1.10)	1070	0.97 (0.91, 1.03)
Military personnel	1470	1.09 (1.03, 1.14)	4	1.01 (0.42, 3.22)
"Other workers"	6597	0.99 (0.97, 1.02)	3541	1.02 (0.99, 1.05)

1.4 Opium

Opium is a highly addictive substance obtained from the unripe seedpod of the poppy plant, and is illicitly consumed by millions of people worldwide, particularly in the Middle East and South Asia ("World Drug Report" 2019). In Western and Central Asia, there is an incorrect belief among laypeople and also among some old-generation physicians that long-term use of opium at a low dose might prevent some chronic diseases such as diabetes mellitus, cardiovascular diseases, and cancer (Kamangar et al. 2014). This wrongly held belief may stem from the analgesic effect of opium which reduces the pain caused by some chronic diseases.

Opium is extracted from the juice (latex) of seeds after special processing to render it appropriate for use (Drug Enforcement Administration (DEA) 1992; Ray, Kattimani, and Sharma 2006). Opium contains a

mixture of alkaloids such as papaverine, codeine, noscapine, morphine, and thebaine (Kalant 1997). Alkaloids might be used as a base for opiate derivates such as heroin (from morphine) and oxycodone (from thebaine) (Yaksh and Wallace 2011). The alkaloid components of opium have beneficial effects including analgesic, hypnotic, antitussive, and antidiarrheal effects (Labanca, Ovesna, and Milella 2018). Opium is often minimally processed by heating, boiling, drying, and variably adulterating with some chemicals (e.g., lead or chromium) before it reaches consumers (Amin-Esmaeili et al. 2016). Opium is used in different forms including teriak (crude/raw opium), shire (refined opium extract), and sukhte (residual opium dross after smoking) (Nasrollahzadeh et al. 2008; Kheirandish et al. 2010). Opium is used via three different methods including oral ingestion, smoking and injection. The injection method is not common. Raw opium and opium juice can be ingested or smoked, while opium dross is usually consumed orally (Masjedi et al. 2013). In addition, opium might be contaminated (intentionally/ unintentionally) during the process of preparing opium from poppy latex into a usable form of opium ("IARC" 2021). Furthermore, accessible opium (street opium) contains some additives such as lead, arsenic, thallium, chromium, cadmium, strychnine, strontium sulfate, starch, and bacteria (Beattie et al. 1975; Etemadi et al. 2022; Hayatbakhsh et al. 2017; IARC Monographs Vol 126 group 2020). Since the type and amount of additives to opium and the kind of opium that is accessible may usually not be standardized products, this variation makes opium a complex agent, complicating the understanding of all aspects of opium and additive mixtures (IARC Monographs Vol 126 group 2020). Therefore, it is important to consider the role of opium additives in cancer risk and compare the opium-cancer association.

Two mechanisms are suggested to underlie the carcinogenic effect of opium consumption, including exposure to opium smoke and the alkaloid component of opium. First, opium users heat the opium to a high temperature and then inhale the smoke through a special pipe when the opium contents vaporize. Opium smoke contains polycyclic aromatic hydrocarbons (PAH), which are confirmed carcinogens (Hewer et al. 1978; Perry et al. 1983). Opium dross can be eaten and may have a mutagenic role after metabolic activation, as shown in Salmonella Typhimurium (Hewer et al. 1978). This effect may be triggered by morphine and opium pyrolysates (Perry et al. 1983). The main carcinogenic components of opium dross consist of nitrogen-containing heterocyclic parts derived from the pyrolysis of morphine (Friesen et al. 1985). In the second mechanism, opium also contains alkaloid compounds that are absorbed in the body and can increase cancer risk (Kamangar et al. 2014).

During the past years, many studies have been carried out to evaluate the carcinogenic effect of opium in humans. These published studies, including both prospective and retrospective studies, evaluated different cancer types including esophageal cancer (Ghadirian et al. 1985; Malekzadeh et al. 2013; Shakeri et al. 2012), gastric cancer (Khosravizadegan et al. 2017; Malekzadeh et al. 2013; Sadjadi et al. 2014; Shakeri et al. 2013), laryngeal cancer (Khoo 1981; Mousavi et al. 2003), lung cancer (Khademi et al. 2012; MacLennan et al. 1977; Masjedi et al. 2013), and BC (Abdolahinia et al. 2021; Akbari et al. 2015; Aliramaji et al. 2015; Asgari et al. 2004; Ghadimi et al. 2015; Hadji et al. 2022; Hosseini et al. 2010; Ketabchi et al. 2005; Lotfi et al. 2016; Nourbakhsh and Hatmi 2006; Rashidian et al. 2021; Sadeghi, Behmard, and Vesselinovitch 1979; Shakhssalim et al. 2010; Sheikh et al. 2020; Toutounchi et al. 2000) (Figure 12). All these studies were conducted in high opium consumption regions and all suggested a positive association between opium use and the risk of cancer (Filho et al., 2023; Kamangar et al., 2014; Mansouri et al., 2022; Singh et al., 2021; Bidary et al., 2021).

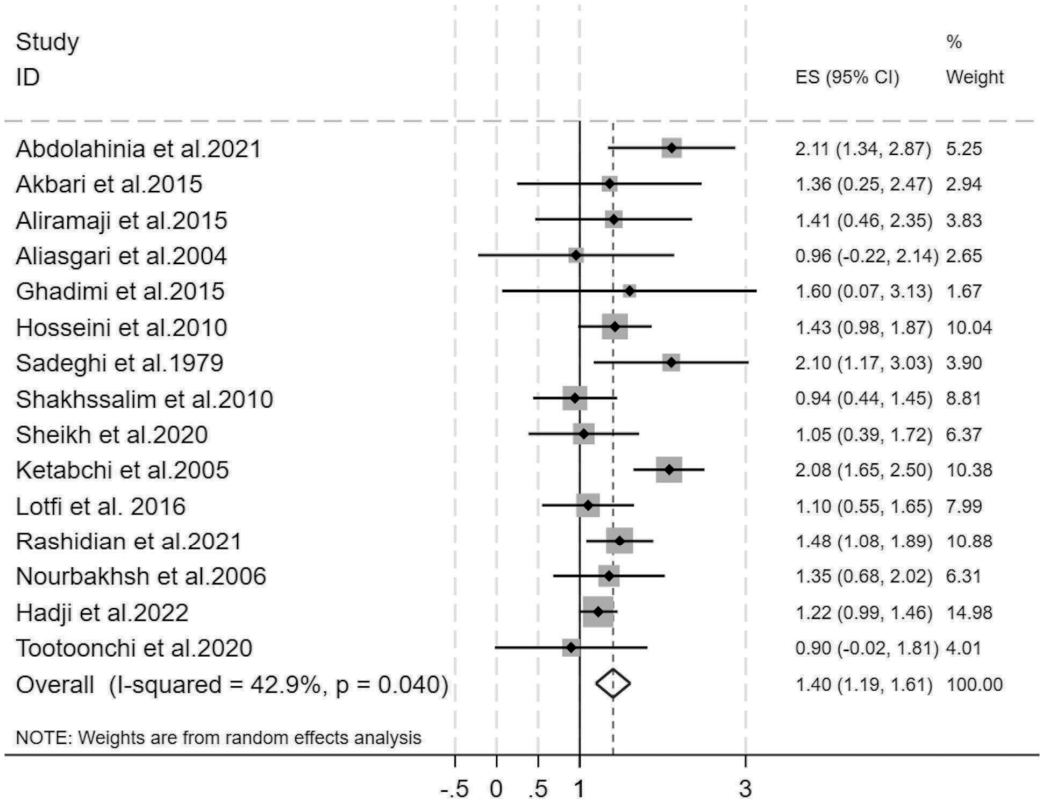

Figure 12. Opium consumption and log relative risk/ odds ratio of bladder cancer according to the published papers.

In September 2020, an IARC Working Group concluded that opium use has a carcinogenic effect on humans based on sufficient evidence of carcinogenicity in humans for cancers of the lung, esophagus, and pancreas (IARC Monographs Vol 126 group 2020). Also, a recent systematic review related to the carcinogenicity of opium consumption reported the overall meta relative risk (mRR) for ever or regular opium use versus never opium consumption in a range of 1.5 for esophageal cancer (mRR: 1.5, 95% CI: 1.1, 2.0) to 8.0 for laryngeal cancer (mRR:8.0, 95% CI: 4.8, 13.3) (Filho et al., 2023). Another systematic review on the carcinogenicity of opium showed a positive association between opium use and the risk of cancer (pooled OR: 3.5, 95% CI: 2.6, 4.8) (Mansouri et al. 2022).

2 JUSTIFICATION OF THE CURRENT STUDY

Most of the studies on cancer risk among opium users have suffered from methodological limitations, such as lack of control for confounding variables, small sample sizes, and lack of information about the starting age of opium use, duration of use, dose, and route of consumption. Therefore, my PhD project helps to improve global knowledge about the carcinogenesis of opium on cancer and supplies further evidence on why opium is a substance that should be banned. The used dataset is unique, containing a large number of cancer cases and controls, and detailed information about opium use and potential confounders collected via a comprehensive, validated questionnaire. The research was initiated six years prior to the classification of opium as a cancer-causing agent by IARC, and the first results were already referred to in the IARC monograph of September 2020. The HNSCC results of this thesis were available to the IARC working group (IARC Monographs Vol 126 group 2020).

3 AIM OF STUDY

The aim of this study is to assess the association between opium use and cancer using the IROPICAN study's dataset collected in Iran. The specific objectives are:

1. To describe the setting and methods of the IROPICAN study.
2. To determine the association between opium use and head and neck, bladder, and colorectal cancers.
3. To determine the role of the duration and dose of opium use in head and neck, bladder, and colorectal cancer risk.
4. To assess the effects of the age of starting and stopping opium use on the association of bladder, and colorectal cancers.
5. To determine the role of the route of opium consumption (smoking, oral use, etc.) on the risk of head and neck, bladder, and colorectal cancers.
6. To determine the interaction of opium and other established risk factors (such as tobacco) with the risk of head and neck, bladder, and colorectal cancers.

4 MATERIALS AND METHODS

4.1 Iranian Study of Opium and Cancer

The Iranian Study of Opium and Cancer (IROPICAN) was launched in 2015 as a large, multi-center case-control study implemented in ten provinces in Iran. These provinces included areas with a moderate to high prevalence of opium consumption (Figure 13). The primary objective of IROPICAN was to investigate the potential association between opium use and the risk of developing lung, colorectal, bladder, and head and neck cancers. The selection of study regions in IROPICAN was based on two primary factors: the prevalence of opium use and the accessibility of cancer care centers for recruiting patients and controls. Before the main study was conducted, a preliminary phase was developed, including a validation study among controls, a validation study among cases, a reliability measurement of self-reported opium use in the questionnaire, and a pilot study.

Figure 13. IROPICAN study provinces in Iran.

4.2 Study population and sampling

This doctoral thesis includes the data of histologically confirmed primary HNSCC, BC, and CRC cases admitted to referral hospitals who were recruited as cases from May 2017 until July 2020 and the pool of all controls of the IROPICAN study. Overall, 919 HNSCC, 717 BC, and 920 CRC were included in this doctoral study.

A crucial aspect of case-control studies is the selection of an appropriate control group, and establishing clear inclusion and exclusion criteria. When selecting hospitalized patients as controls, it is important to ensure that they do not have the disease of interest and that their current condition is not associated with the exposure of interest. In other words, the controls should be chosen from a population that is at risk of developing the disease but has not yet developed it, and their exposure status should be representative of that of the population from which the cases were drawn (Lewallen and Courtright 1998; Schulz and Grimes 2002). In addition, the frequency of exposure in the control group should be representative of the exposure frequency in the population from which the cases were extracted (Grimes and Schulz 2005). While population-based controls are a better option for case-control studies, they may not always be feasible or appropriate for certain situations. For example, when studying an association between cancer and illegal drug use, population-based controls may underreport their exposure history compared to cancer patients, leading to differential misclassification and overestimation of the disease-exposure association. In such cases, alternative methods for control selection, such as hospital-based controls, may be more appropriate to minimize bias and accurately assess the association between the disease and exposure of interest.

Hence, in June and July 2016, a validation study was conducted by the IROPICAN team to determine the optimal selection procedure for the control group and to estimate the size of underreporting bias. The study evaluated whether hospitalized individuals with diagnoses other than cancer or hospital visitors would be a better source of control group (Rashidian et al. 2017).

In the validation study for the hospitalized patients' group, purposive (convenience) sampling was utilized to select hospitals and wards for the study. Specifically, the hospitals that were chosen were referral centers for patients located in each province. Subsequently, hospitalized patients with a diagnosis unrelated to the study's main exposure of interest (i.e., opium use) and who received treatment at these hospitals were recruited for the study. This approach allowed the IROPICAN team to recruit patients who had a referral pattern resembling that of cancer patients. To ensure that the hospitalized patient group resembled the cancer patients more closely, both chronic and acute patients were recruited. Then, hospitalized patients were selected using stratified random sampling based on a five-year age interval and the patient's place of residence (whether they lived in or outside of the capital city of their province). Patients who were too ill to complete the questionnaire were not included in the study. On the other hand, a sampling method similar to that used for hospitalized patients was employed for the group of healthy individuals, and individuals who were accompanying patients in the same hospitals were selected.

To validate the self-reported opium use, we used two types of urine drug screening tests, urine rapid drug screen (URDS) and immunoassay thin-layer chromatography (TLC). The former is one of the most commonly used methods for initial screening. The latter is a confirmatory testing method that uses antibodies to detect the presence of specific drugs or their metabolites. It is considered the gold standard for drug testing due to its high sensitivity, accuracy, and reliability. TLC can detect even small amounts of

a substance and is the most accurate method of testing. However, TLC is a time-consuming process that requires a high level of expertise to perform, and it can be costly. For these reasons, TLC is typically done only after a positive result is obtained in the URDS test. The URDS test is a quick and less expensive method of drug screening that can be used as an initial screening procedure before more confirmatory testing is performed using TLC (Moeller, Lee, and Kissack 2008).

For the initial screening, the study used the URDS test to assess the presence of opium metabolites, specifically morphine, with a cutoff point of 300 ng/ml. The test protocol was developed by a toxicologist from the Iranian National Center for Addiction Studies (INCAS). Metabolites of morphine are typically detectable in urine for up to 72 hours after use (Matson 2011). Therefore, TLC tests were used for participants who tested positive in the URDS test but denied either regular use of opium or the use of the drug during the previous 72 hours in the questionnaire. The purpose of the TLC test was to confirm the presence of opium metabolites and rule out false positives from the URDS test.

The participation rate was high in both healthy individuals (86%) and hospitalized patients (88%). Non-participants were defined as people who refused to participate after receiving information about the study. Approximately half of the non-participation rates in both groups were due to the person's unwillingness to provide urine samples. Overall, the high participation rates suggest that the study was well-received by the participants and that the sampling method was effective in recruiting a representative sample of both healthy individuals and hospitalized patients.

The under-reporting rate was 25% for the disease control group and 30% for the healthy visitors (P-value > 0.4). In terms of opium use prevalence among the validation study participants, 36% (95% CI: 28%, 43%) of the disease control group and 19% (95% CI: 13%, 25%) of healthy visitors reported having used opium at least once a week for six consecutive months during their lifetime. The study results indicated that healthy visitors were a more suitable control source for this case-control study (Rashidian et al. 2017).

Likewise, the IROPICAN team conducted a validation study to investigate the potential differences in self-reported opium use between cancer patients and patients with other diseases. For this purpose, 100 cancer patients were selected from those who were candidates for surgery at the Cancer Institute of Iran, located in Tehran. The individuals in this phase were selected from the patients with different forms of cancer. To ensure that the findings could be compared with those obtained in the control group validation study, only male participants were included. During the interview process, information on the participants' history of opium use and demographic factors was collected, and subsequently compared to the anesthesia report. It is standard practice for anesthesiologists to ask about a patient's opium use before surgery to prevent withdrawal symptoms. Since many cancer patients use opioids for pain management, it was not feasible to use urine tests as a reliable measure. Instead, the patient's self-reported opium use as documented in the anesthesia report was considered the gold standard. The mean age of the study participants was 61.5 years (±SD: 16.3). The prevalence of opium use among the participants was 18% (95% CI: 11.7%, 26.7%). The sensitivity of self-reported opium use was found to be 69.6% (95% CI: 49.1%, 84.4%).

4.3 Case recruitment

Cancer patients who had been diagnosed with primary cancer of the colorectum (C18-21), bladder (C67), or squamous cell carcinoma (SCC) of the head and neck, specifically in the oral cavity (C03-06), larynx (C32), or pharynx (C10-C14) were recruited to this study. These patients admitted to referral hospitals of the medical universities of ten provinces (Tehran, Shiraz, Kerman, Golestan, Mazandaran, Kermanshah, Khorasan-Razavi, Bushehr, Hormozgan, and Systan-Balouchestan; Figure 13) were enrolled in the study as cases. The cancer cases were individuals who had been diagnosed with cancer within the previous year and had resided in the study regions for at least two years. To be eligible for inclusion in the study, participants had to meet several criteria, including being of Iranian nationality, having an ability to speak and understand Persian (Farsi), being willing and able to participate in an 80-minute interview, and being between the ages of 30 and 75 years. Patients with metastatic cancer, a second primary cancer, or without a confirmed pathology report were excluded from the study, as were pregnant and nursing women.

4.4 Control recruitment

Controls were selected at the same time as the cases and had to meet certain criteria to be included in the study. They had to be between the ages of 30 and 75 years and free of any type of cancer. Controls were selected from hospital visitors, including the relatives or friends of patients from non-oncology wards as well as other individuals who visited the hospital for reasons other than receiving treatment. All controls were frequency matched with the cases based on their gender, place of residence, and age, with age intervals of five years. The inclusion criteria for controls were the same as those for cases in terms of age, nationality, ability to speak and understand Farsi, and pregnancy/nursing status. Control subjects had to be between the ages of 30 and 75 years, of Iranian nationality, able to speak and understand Farsi, and not pregnant or nursing. Controls for each cancer type were selected separately. The controls were selected every three months after the recruitment of a sufficient number of cases and were frequency-matched for age (5 years age range), sex, and place of residence for each cancer type. They were healthy visitors who came to the general hospital. In final analyses, the control pool of controls of all cancer types combined was used for each cancer type, and the age, region and gender distribution of controls therefore differed from that of the cases. This imbalance was taken into account in the analyses and did not cause bias in the ORs.

4.5 Information on exposure

In the study, the ever-use (lifelong) of opium was defined as having used opium at least once during a person's lifetime. Meanwhile, regular opium use was defined as having used opium at least once a week for a minimum of six consecutive months.

The detailed histories of regular opium use included the duration of opium use, starting and stopping age, and the amount and frequency of opium use per day, week, and month. The amount of opium use was measured in local units of opium use (i.e., nokhod, hab, dood, adas, mesghal), which were later converted to the standard unit, i.e., grams.

The type of opium used, including crude or raw opium, opium juice or shireh (i.e., a refined condensed extract of the remnants of smoked opium (Amin-Esmaeili et al. 2016)), and sukhteh (i.e., remnants of

smoked opium), as well as the routes of administration (smoking, ingestion) were also recorded. Participants were asked to provide responses for up to five separate periods of opium use.

Similar to the collection of opium use history, the study also gathered detailed information on other exposures, such as cigarette smoking and tobacco use, and alcohol intake. Specifically, the use of various forms of tobacco, including waterpipe, nass, chopogh, and pipe, was recorded. Ever-use (lifelong) of cigarettes and tobacco (including waterpipe, chopogh, nass, and pipe) was defined as having used any of these at least once during one's lifetime. Regular cigarette smoking and tobacco use were defined as using any of these at least once a week for a minimum of six consecutive months. Likewise, lifetime alcohol intake was defined as having consumed alcohol at least once during one's lifetime. Regular alcohol intake, on the other hand, was defined as having consumed alcohol at least once a week for a minimum of six consecutive months.

Another form of exposure examined was physical activity. In this study, we utilized the perceived physical workload (PPWL) of participants. To estimate the PPWL of participants, the study used the Finnish Job Exposure Matrix (FINJEM) and all of the participants' job histories. The PPWL was determined based on three factors: the participant's occupation, the proportion of exposure (P), and the mean level of exposure among those exposed (L) during a specific calendar time. The study also gathered information on the participants' body shapes by having them complete a pictogram questionnaire at different ages including one year before the interview time, at the age of 15, and the age of 30 years. The study also recorded the participants' family history of cancer, including any instances of cancer among their first-degree relatives. The study also collected detailed information on the participants' education level (measured as years of education), as well as their ownership of certain assets, including a vacuum cleaner, washing machine, dishwasher, freezer, internet access, microwave, laptop, mobile phone, personal car, and shop. All this information defined the socio-economic status (SES) of participants.

Additionally, the study collected information on the participants' nutrition using a validated Persian cohort food frequency questionnaire (FFQ), which measured 131 different food items (Poustchi et al. 2018). The participants were asked to complete the FFQ for the year preceding their interview.

4.6 Questionnaire

A detailed questionnaire adapted from the Golestan Cohort Study (Pourshams et al. 2010) was administered to all cases and controls. The questionnaire included questions on age, ethnicity, education, rural/urban status, occupational history, SES, physical activity, oral health, female reproductive history, disease history, personal and family history of cancer, tobacco consumption (cigarette and water-pipe, pipe, nass, and chopogh), lifelong alcohol consumption, opium use as well as other drug use, use of opiates such as non-medicinal morphine, raw opium, shireh (i.e., a refined condensed extract of remnants of smoked opium (Amin-Esmaeili et al. 2016)), sukhteh (i.e., remnants of smoked opium), and other drugs including heroin, crack of heroin, hashish, tramadol, methadone, etc. Additionally, a validated and reproducible 131-item food frequency questionnaire (FFQ) (Poustchi et al. 2018) was used to collect dietary information.

4.7 Reliability of the questionnaire

The reliability of the questionnaire was assessed for the questions related to opium and tobacco use, drug abuse, tobacco use, and alcohol use. In this step 57 participants were recruited to study from the addiction control center. This phase was conducted at Methadone Maintenance Treatment (MMT) centers which are registered centers that offer treatment programs to drug addicts. The subjects for this study phase were selected based on inclusion criteria which were as follows: the subject should a) be Iranian, b) be able to answer the questionnaire in 15 minutes, c) have a possibility to be referred to a re-interview carried out after two weeks, d) be able to understand and communicate with the interviewer, e) be aged between 25 to 65 years old, f) attend treatment for any illegal drugs except alcohol, g) have used opium or any drugs during the previous six months. The main reason for implementing this study in the MMT center was the fact that this would provide us with access to individuals and a follow-up carried out two weeks later. The reliability of questionnaire responses was assessed using an intra-class correlation coefficient (ICC) that involved comparing the subjects' first and second responses. We used ICC because this index measures both the degree of correlation and the agreement between measurements (Koo and Li 2016).

Out of all the participants, 47 (82.5%) were men. The median age was 21 (range 10–41) years old for starting cigarette use and 21 (range 17–25) for starting alcohol use. Meanwhile, the median starting age of opium use was 24 (range 13–50) years old. The lifetime duration of cigarette, alcohol and opium use was 21, 12, and 13 years, respectively. The Cronbach's Alpha of the questionnaire was 0.97.

The ICC of regular cigarette use was 0.95 (95% CI: 0.94, 0.97). Moreover, the ICC of regular use of flavored water pipe, non-flavored water pipe, and chopogh was 0.95, 0.99, and 0.99, respectively. In addition, the ICC of lifelong opium use was 1.0 and the ICC of regular use of opium was 0.88 (95% CI: 0.80, 0.92) (Table 5). Moreover, The ICC of regular opium use for men was 0.88 (95% CI: 0.80, 0.93) and for women 0.88 (95% CI: 0.60, 0.97) (not shown in the tables).

Table 5. The intraclass correlation coefficient (ICC) for the lifetime and regular use of different drugs, tobacco, and alcohol use.

Variables	ICC	CI (95%)
Lifetime cigarette use	0.96	(0.94, 0.97)
Regular cigarette use	0.95	(0.91, 0.97)
Regular flavored water pipe use	0.99	(0.98,0.99)
Regular non-flavored water pipe use	0.99	(0.98, 0.99)
Regular chopogh use	0.96	(0.93, 0.98)
Lifetime tobacco use *	0.88	(0.80, 0.93)
Lifetime opium use	1	
Regular opium use	0.88	(0.80, 0.92)
Lifetime heroin use	0.95	(0.92, 0.97)
Regular heroin use	0.92	(0.86, 0.95)
Lifetime shisha use	1	
Regular shisha use	1	
Lifetime hashish use	0.98	(0.96, 0.98)
Regular hashish use	1	
Lifetime codeine use	0.89	(0.81, 0.93)
Regular codeine use	0.94	(0.89, 0.96)
Lifetime tramadol use	0.90	(0.82, 0.94)
Regular tramadol use	0.84	(0.72, 0.91)
Lifetime diphenoxylate use	0.95	(0.92, 0.97)
Regular diphenoxylate use	0.84	(0.71, 0.91)
Lifetime methadone use	0.94	(0.90, 0.96)
Regular methadone use	0.97	(0.96, 0.98)
Lifetime alcohol use		
Regular alcohol use	0.99	(0.98, 0.99

*Flavor water pipe, non-flavor water pipe, and chopogh

Based on the responses, the most common method of opium use was the ingested method 43%. The ICC of this opium use method was 0.78 (95% CI: 0.63, 0.90). Opium users may apply different methods of opium use, which can vary according to the type of opium, the use environment (indoors or outdoors), and other diseases the person might have. As a result, the users may be unable to recall the methods of use. That is why the ICC for the opium method is low. Of the people who had used opium, 70% had used crud opium and the ICC of opium type was 0.97 (95% CI: 0.95, 0.98) (Table 6).

Table 6. The intraclass correlation coefficient (ICC) and 95% CI for the duration and type of different drug, tobacco, and alcohol use.

Variables	ICC	CI (95%)
Duration of cigarette use	0.96	(0.94, 0.98)
Duration of flavored waterpipe use	1	
Duration of non-flavored waterpipe use	0.90	(0.83, 0.94)
Duration of chopogh use	1	
Starting age for opium use	0.94	(0.92, 0.97)
Duration of opium use	0.89	(0.81, 0.93)
Type of opium	0.97	(0.95, 0.98)
Way of opium use	0.78	(0.63, 0.90)
Total alcohol use amount	0.99	(0.98, 0.99)
Type of alcohol	0.99	(0.98, 0.99)
Duration of alcohol use	0.98	(0.98, 0.99)
Passive exposure to cigarette	0.98	(0.96, 0.98)
Passive exposure to opium	0.91	(0.84, 0.94)

4.8 Pilot study

Finally, a pilot study was conducted from January to April 2017. Patients with four types of cancer were recruited as the case group, while healthy individuals visiting referral hospitals and clinics in four different provinces of Iran (Tehran, Shiraz, Kerman, and Golestan) were recruited as the control group. The main objectives of the pilot study were to assess the feasibility of conducting a case-control study on cancer in Iran, optimize the questionnaires used to collect data from participants, provide training to partner research institutions for data collection, evaluate geographic variation in opium use and other opiates across different regions of Iran, and optimize the methods used to collect biological samples from study participants.

4.9 Statistical analysis

In order to evaluate the association between opium use and the risk of cancer, both univariate and multivariable unconditional logistic regression models were utilized to determine crude and adjusted odds ratios (ORs) and 95% confidence intervals (CI). An adjustment for potential confounders for each type of cancer and opium was included, in addition to matching the factors of age, gender, and place of residence. To account for reverse causality bias, a three-year lag time was implemented, which excluded opium consumption during the three years preceding the interview date.

The information about the amount of opium consumed daily was reported using different local units. To standardize the opium unit, all local measurement units were converted to grams, as shown in Table 7. In addition to opium use, the participants were asked about their use of other drugs, including heroin, methamphetamine, cannabis, and prescription opioids, such as diphenoxylate, codeine, tramadol, and methadone.

Table 7. Conversion of traditional units of opium to grams (International System of Units).

Unit	Grams
Nokhod	0.2
Bast	0.1
Mesghaal	4.58
Hab/habeh	0.2
Dood	0.07
Adas	0.048
Spoon	5
Milliliter	1

Opium use was categorized into ever-use or regular use. Ever-use was defined as using opium at least once over the participant's lifetime, while regular use was defined as using opium at least once a week for at least six consecutive months. To determine the opium amount (in grams) and frequency of opium use (per day), it is necessary to take into account the duration of opium use as it varies among users. Therefore, the calculation of the weighted average of opium amount and frequency should consider the duration of opium use.

The daily dose of opium is calculated by multiplying the weighted average of opium use and the weighted average of times per day. Consequently, lifelong opium use throughout one's lifetime is determined by multiplying the weighted average of opium use, the weighted average of times per day, and the total duration of opium use. In all analyses, the group of non-users is used as the reference group.

As opium is an illegal drug, it is unlikely that its use was measured perfectly accurately for all participants, and there may have been some degree of error in the measurements. A validation study (Rashidian et al. 2017) showed that the sensitivity (Sn) of the self-reporting of opium use was 70% among cases and 69% among controls. Moreover, the specificity (Sp) of the self-reporting of opium use among both cases and controls would be 100%. This suggests that any potential bias in the study results would likely be towards the null. To address the issue of non-differential misclassification, a quantitative bias analysis was performed using probabilistic bias analysis. This involved applying multiple combinations of Sn and Sp in a multidimensional correction, and cross-tabulating the corrected associations to compare the relative impact of different assumed values.

The use of tobacco (such as cigarette smoking, waterpipe use, pipe use, nass, and Chopogh) was considered the primary confounder that needed to be taken into account for HNSCC and BC. To determine the confounding effect of tobacco use on the association between opium use and cancer, both multiplicative and additive interactions were evaluated. The interaction contrast was assessed using measures such as the relative excess risk due to interaction (REIRI), the attributed proportion of outcome among those with both tobacco smoking and opium use that is attributed to their interaction (AP), and the synergy index (S) (Andersson et al. 2005).

We created an SES variable by conducting principal component analyses on data related to the participants' years of education (a continuous variable) and their ownership of various items, including a vacuum cleaner, washing machine, dishwasher, freezer, microwave, laptop, internet access, mobile phone, personal car, and a shop. Using tertiles derived from the principal component analysis, we then categorized SES into three groups: low, moderate, and high.

In study II, we decided to conduct a multilevel logistic regression. As we recruited participants from ten different centers, we tested for heterogeneity between the centers (P-heterogeneity) and applied mixed

effects logistic regression models with random intercepts by the center of study to estimate the association between opium use and HNSCC status (odds ratio with 95% confidence interval).

The potential confounders for HNSCC included age, gender, place of residence (urban/rural), cigarette smoking (measured in pack-years), water-pipe smoking (measured in head-years), alcohol drinking (regular drinkers vs non-regular drinkers), oral health (measured using the DMFT index), and SES. Analyses were conducted for all HNSCCs, as well as for the four main anatomic sub-sites such as lip and oral cavity including codes C00-C08 and C14, pharynx codes C09-C11 and C13, larynx codes C32, and other sub-sites within head and neck codes C12, C31, C32, and C76. Heterogeneity between the centers was also tested and a mixed-effects logistic regression model with random intercepts by the center of the study was applied.

Furthermore, as tobacco and alcohol are the two identified risk factors for HNSCC, a subgroup analysis was also performed, limited to individuals who had never smoked tobacco.

In study III, the potential confounders included the pack-years of cigarette smoking, age, gender, and place of residence. Occupation as an important confounder for BC was dropped from the model due to the fact that it did not improve the model fitness (P-value >0.2)

To assess the association between opium consumption and the risk of CRC (study IV), the models were adjusted for various potential confounding variables, such as age, gender, province, marital status, SES, body shape, perceived physical workload (PPWL), family history of cancer, red meat intake, and vegetable consumption.

Another variable was the family history of cancer. Those who reported one or more cancers of the colon, rectum, stomach, ovaries and endometrium in their first-degree relatives were defined as having a positive family history of cancer. These cancer types are possibly linked to Lynch syndrome or hereditary nonpolyposis colorectal cancer (HNPCC). HNPCC is an autosomal dominant genetic condition that is associated with a high risk of colon cancer and cancers of the endometrium (second most common), ovary, stomach, small intestine, hepatobiliary tract, upper urinary tract, and brain, and skin cancers (Hampel et al. 2005; Peltomäki, Olkinuora, and Nieminen 2020). None of the participants reported a history of hepatobiliary tract, upper urinary tract, brain, or skin cancers in their first-degree relatives.
Using the job histories of the participants and the Finnish Job Exposure Matrix (FINJEM), the PPWL was calculated (Kauppinen et al. 2014; T. Kauppinen, Toikkanen, and Pukkala 1998). FINJEM contains two variables, namely the proportion of exposed (P) and the mean level of exposure among the exposed (L), which are utilized to estimate the PPWL for each occupation and calendar period in FINJEM (Pukkala et al. 2005; Kauppinen, Toikkanen, and Pukkala 1998). The estimated cumulative PPWL was calculated by multiplying the P, L, and the years worked in the job with the longest duration for each person. However, due to overlapping work periods in the interview data, it was not possible to calculate the cumulative PPWL for the persons' entire work history. The cumulative PPWL was categorized into three groups: sedentary (zero PPWL-years), moderate (PPWL-years greater than zero and less than or equal to 4.80), and heavy (PPWL-years greater than 4.81).

Also, to calculate the daily intake of each food item, the reported daily frequency of consumption was multiplied by the standard portion size in grams. Subsequently, red meat, fruit, and vegetables were categorized into three groups using tertiles as cut-off points.

Moreover, body shape was used based on a pictogram questionnaire that the participants used to report their idea of their body shape when they were 15 years old. To assess the participants' perception of their body size, a set of pictograms was used ranging from very lean to obese. The pictogram was scored between 1 to 7 in males and 1 to 9 in females. The validity of using pictograms to distinguish between obese, overweight or normal-weight participants was explained in detail elsewhere (Keshtkar et al. 2010). By using this pictogram, it was possible to determine whether the participants' body mass index (BMI) suggested they were obese (BMI ≥30), overweight, (BMI 25-<30), or normal (BMI18.5-<25) and to categorize them into these three groups. Participants scored the pictogram in three different time scales including one year before the interview date, at the age of 15 years, and at the age of 30 years. Finally, the most appropriate age was selected to estimate their BMI. The scores of one to three were defined as normal weight, four as overweight, and five to nine as obesity.

To avoid overadjustment bias due to the adjustment of the mediator, all potential confounders were adjusted but intermediate (mediator) variables such as constipation and oral hygiene were not. Mediators are variables that alter the effect of exposure on the outcome through a specific mechanism. Opium always contains additives like lead (Beattie et al. 1975; Hayatbakhsh et al. 2017; "IARC" 2021; Masoudi et al. 2006), and lead can cause constipation (Sundbøll et al. 2019). On the other hand, for those who suffer from constipation, the risk of CRC is higher (Sundbøll et al. 2019). Therefore, based on causal directed acyclic graphs (cDAGs) (Etminan, Collins, and Mansournia 2020), it was concluded that constipation plays an intermediate role between opium consumption and the risk of CRC, not as a confounder (Figure 21). Along with constipation, there is a association between oral health and the risk of CRC: a low oral health index might increase the risk of CRC (Momen-Heravi et al. 2017). On the other hand, opium use is associated with periodontal disease (Wu et al. 2021). Consequently, oral health was considered the mediator between opium use and the risk of CRC (Figure 14).

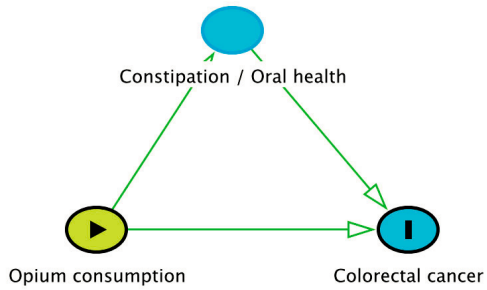

Figure 14. Causal-directed acyclic graph for opium use and colorectal cancer risk with constipation/oral health as a mediator.

In addition to the DAGs, which show the intermediate variables, we also conducted a mediation analysis to prove the intermediate factors such as constipation and oral hygiene statistically. We performed a series of mediation analyses (VanderWeele 2015) in which these risk factors were regressed on opium use to clarify their role in explaining the association between opium use and the risk of CRC.
The mediation model was based on standard approaches for binary mediator and outcome, comprising two logistic regressions, one for CRC risk, which allows for potential interaction between opium use and the other risk factors:

logit {P (Y=1 | d, m, c)} =00+01d+02m+03dm+0'4c

and one for mediators:

logit {P (M=1 | d, c)} = β0 + β1d + β'2c

where D denotes the exposure of interest (opium use), M the mediators (constipation, oral hygiene), Y the outcome (CRC), and C a set of potential confounders (e.g., gender, age, race/ethnicity, year of enrolment) (VanderWeele and Vansteelandt 2014). The relationship between these factors is depicted in Figure 15. Assuming no residual confounding the OR for the direct effect (i.e., the effect of opium use and CRC risk that remains after allowing for the effect of potential mediators) and the indirect effect (i.e., the effect on the association between opium use and the risk of CRC that is explained by the mediators) will be obtained using the formulae mentioned above (VanderWeele, 2015).

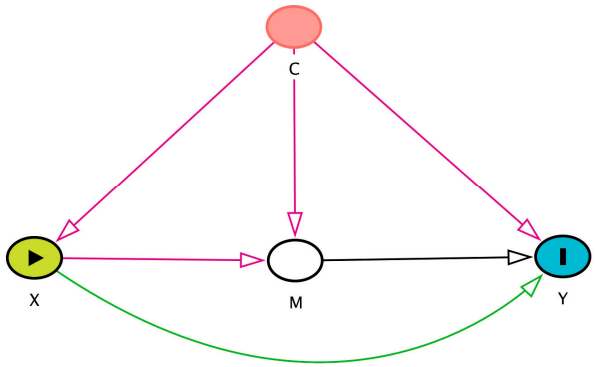

Figure 15. Graphical representation of a mediation analysis

4.10 Software

All statistical analyses were conducted using Stata, version 17 (Stata Corp, College Station, Texas 77845 USA, licensed to Tampere University).

4.11 Ethical Consideration

The study received approval from the Institutional Review Boards of the National Institute for Medical Research Development (NIMAD), Iran, under the reference number IR.NIMAD.REC.1394.027. Prior to collecting data, written consent was obtained from each study participant. To protect the privacy of the participants, their information was de-identified through unique identification codes and kept in databases with restricted access.

5 RESULTS

5.1 Characteristics of the study population

5.1.1 Study I: describe the methods of the IROICAN study and present baseline findings.

The study involved 3,289 individuals who had been diagnosed with cancer (740 with lung cancer, 914 with CRC, 717 with BC, and 918 with HNSCC) and 3,477 individuals who did not have cancer and served as controls. Among the individuals who had been diagnosed with cancer, 98% responded to the study (3,247 out of 3,314), while among the control group, 87% responded (3,397 out of 3,925). Of all the participants, 73% of the cancer cases and 69% of the controls were male (Table 8).

Table 8. Demographic characteristics of cases enrolled in the multi-center case-control study of opium and cancer (IROPICAN study), 2015–2019, by cancer site.

Variable		Site				Controls
		Lung N (%)	Colon and rectum N (%)	Bladder N (%)	Head and neck N (%)	
Total		740 (100)	914 (100)	717 (100)	918 (100)	3477 (100)
Age						
	30-39	24 (3.2)	67 (7.3)	14 (2.0)	64 (7.0)	257 (7.4)
	40-49	91 (12.3)	131 (14.3)	50 (7.0)	144 (15.7)	559 (16.1)
	50-59	230 (31.1)	256 (28.0)	181 (25.2)	277 (30.2)	1070 (30.8)
	60-69	255 (34.5)	274 (30.0)	267 (37.2)	295 (32.1)	1092 (31.4)
	≥70	140 (18.9)	186 (20.4)	205 (28.6)	138 (15.0)	499 (14.4)
Gender						
	Female	79 (24.2)	387 (42.3)	93 (13.0)	232 (25.3)	1077 (31.0)
	Male	561 (75.8)	527 (57.7)	624 (87.0)	686 (74.7)	2400 (69.0)
Province						
	Tehran	148(20.0)	170 (18.6)	139 (19.4)	163 (17.8)	816 (23.5)
	Fars	233 (31.5)	265 (29.0)	166 (23.2)	379 (41.3)	943 (27.1)
	Kerman	128 (17.3)	105 (11.5)	150 (20.9)	160 (17.4)	525 (15.1)
	Golestan	64 (8.7)	156 (17.1)	46 (6.4)	46 (5.0)	374 (10.8)
	Mazandaran	36 (4.9)	59 (6.5)	24 (3.4)	17 (1.9)	136 (3.9)
	Kermanshah	42 (5.7)	71 (7.8)	52 (7.3)	37 (4.0)	251 (7.2)
	Khorasan-Razavi	16 (2.2)	88 (9.6)	30 (4.2)	44 (4.8)	170 (4.9)
	Bushehr	38 (5.1)	.*	56 (7.8)	.**	84 (2.4)
	Hormozgan	17 (2.3)	.*	27 (3.8)	36 (3.9)	78 (2.4)
	Sistan-Baluchestan	18 (2.4)	.*	27 (3.8)	36 (3.9)	100 (2.9)

*Colorectal cancer cases were not collected in Bushehr, Hormozgan, and Sistan-Baluchestan

**Head and neck cancer cases were not collected in Bushehr

Of the individuals who chose not to participate in the study, 66% of those diagnosed with cancer and 71% of the controls were male. Meanwhile, among the participants of the study, the mean age of those diagnosed with cancer was 59.7 years (±SD: 11.9), and the mean age of those who did not participate was

58.8 years (±SD: 11.2). The controls who participated in the study had a mean age of 57.1 years (±SD: 11.6), whereas those who did not participate had a mean age of 58.3 years (±SD: 10.9). The primary reason for not participating among the cancer patients was sickness and lethargy, which accounted for 93% of the non-participants. Meanwhile, among the controls, the primary reason for non-participation was either lack of time or unwillingness to provide a biological sample, which accounted for 83% of the non-participants.

The study revealed that 40.2% of the individuals diagnosed with cancer and 17.4% of the controls had a history of opium use throughout their lifetime. It is possible that the patients started using opium as a way to alleviate their pain, which may have been an early symptom of the cancer that was later diagnosed. This could lead to reverse causality, where opium use seems to cause cancer when in fact the cancer caused the opium use. When individuals who started using opium within three years of their interview date were considered non-users, the study still identified 35.4% of the cancer patients and 12.6% of the controls as opium users.

The median and interquartile range (IQR) of opium use throughout the lifetime of users were 0.8 grams per day (with an IQR of 2.8) for the cancer patients and 0.4 grams per day (with an IQR of 1.9) for the controls. The prevalence of regular opium use varied across different provinces, with the highest prevalence being observed in the eastern regions (including Kerman, Sistan-Baluchestan, and Khorasan-Razavi), where 37% of the cancer patients and 43.0% of the controls reported regular opium use (Table 9).

Among the opium users, there were differences in the types of opium consumed. Specifically, 78.1% of the cancer patients and 88.5% of the controls reported using crude opium. The most commonly reported method of opium consumption was smoking alone, with 66.3% of opium-using cancer patients and 83.9% of controls reporting this method. Ingestion mixed with smoking was another frequently reported method (Table 9).

The study found that the lifetime prevalence of water-pipe use was 18.1% among the cancer patients and 14.4% among the controls. More than half of these individuals reported being regular water-pipe users. The study also revealed that the lifetime prevalence of cigarette use was higher among cancer patients, with 51% reporting having smoked cigarettes compared to 31% among the controls. Among those who had smoked cigarettes at any point in their lives, the majority reported being regular smokers, with 92.2% of the cancer patients and 83.1% of the controls reporting regular cigarette use (Table 9).

Table 9. Characteristics of the cases and controls in the multi-center case-control study of opium and cancer (IROPICAN study), 2015–2019.

Variable	Cases N (%)	Controls N (%)
Total	3247 (100)	3397 (100)
Opium use		
Never	1941 (59.8)	2802 (82.5)
Ever users	1305 (40.2)	592 (17.4)
Non-regular use	119 (3.7)	150 (4.4)
Regular use	1186 (36.5)	442 (13.0)
Not known	1 (0.0)	3 (0.1)
Out of regular opium users	1186 (100)	442 (100)
Region		
Center	155 (13.1)	72 (16.3)
Southern	384 (32.4)	82 (18.6)
Eastern	439 (37.0))	191 (43.2)
Northern	147 (12.4)	63 (14.3)
Western	61 (5.1)	34 (7.7)
Median opium amount		
≤ 0.4 gram/day	327 (27.6)	201 (45.5)
>0.4 gram/day	859 (72.4)	241 (54.5)
Opium use duration		
≤ 17 years	434 (36.6)	230 (52.0)
> 17 years	752 (63.4)	212 (48.0)
Initiation age of opium use		
≤30 years	592 (49.9)	222 (50.2)
>30 years	516 (43.5)	183 (41.4)
Unknown	78 (6.6)	37 (8.4)
Type of opium use		
Crude opium (Teriak)	926(78.1)	391 (88.5)
Opium juice (Shireh)	88 (7.4)	26 (5.9)
Opium dross (Sookhteh)	5 (0.4)	1 (0.2)
Mixed method	167 (14.1)	24 (5.4)
Route of opium use		
Only smoking	786 (66.3)	371 (83.9)
Only ingestion	177 (14.9)	39 (8.8)
More than one route	219 (18.5)	31 (7.0)
Unknown	4 (0.3)	4 (0.2)
Waterpipe use		
Never	2659 (81.9)	2908 (85.6)
Ever users	588 (18.1)	489 (14.4)
Non-regular use	240 (7.4)	211 (6.2)
Regular use	348 (10.7)	278 (8.2)
Cigarette use		
Never	1592 (49.0)	2274 (66.9)
Ever users	1655 (51.0)	1123 (33.1)
Non-regular use	129 (4.0)	190 (5.6)
Regular use	1526 (47.0)	933 (27.5)

According to the pilot study results, some minor revisions were made to the questionnaire, with a focus on improving questions related to drug use. Based on the focus of the study on opium use as the main exposure, the questionnaire included detailed questions related to opium use. Therefore, to improve the accuracy of data on opium use, the questions related to opium use were formulated in greater detail, and a question about the frequency of opium use per day, week, month, or year was added to allow for more precise calculation of the dose.

In the pilot study, the aim was to find frequency-matched controls based on age, gender, and place of residence, but due to time limitations, this could not be achieved. However, in the main study, better frequency matching of the control group was achieved, as more time and resources were allocated for data collection, and the sample size was larger. Based on the experiences gained from the pilot study, only minor changes were made to the sample collection and methodology when the full project was set up.
Assuming a 20% prevalence of opium use in the high prevalence regions of Iran, a power of 90% and a significance level of 0.05 for identifying an odds ratio of 2.0, based on calculations, a decision was made to recruit a sample size of 250 cases and 250 controls. However, the study decided to increase the sample size to 3,200 cancer cases (i.e., 800 cases for each type) and 3,200 common controls to allow for subgroup analyses.

5.1.2 Study II

To investigate the association between opium use and HNSCC, a total of 663 HNSCC cases (254 lip and oral cavity, 54 pharynx, 327 larynx, and 28 other sub-sites within head and neck) and 3,065 frequency-matched controls were enrolled in the study. Table 10 presents information about how demographic and habitual factors are distributed between the two groups of cases and controls. The data indicate that a higher proportion of cases were male (about 75%) and lived in urban areas (about 73%) compared to controls, where the corresponding percentages were 68% and 78%, respectively. Additionally, the median age at recruitment was slightly higher for cases at 58 years old (with a range of 50 to 66 years old for 25% to 75% of cases), whereas it was 57 years old for controls (with a range of 49 to 64 years old for 25% to 75% of controls). Individuals classified as cases had a higher likelihood of smoking, drinking alcohol, having lower SES, and poorer oral health compared to controls (Table 10).
Of the cases, two per cent and of the controls, 14% did not respond. The non-responses for both cases and controls were mostly due to refusals, with the primary reason being unwillingness to provide blood samples and other unspecified reasons. There was no observable variation in age and gender between the individuals who participated in the study and those who did not respond.

Table 10. Distribution of demographics and habits for head and neck squamous cell carcinoma (HNSCC) cases and controls.

	HNSCC cases [a] N (%)	Controls N (%)	P
Total	663	3065	
Age			0.360
≤29	9 (1.4)	25 (0.8)	
30-39	45 (6.8)	246 (8)	
40-49	100 (15)	517 (16.9)	
50-59	213 (32.2)	982 (32.1)	
60-69	203 (30.6)	923 (30.1)	
≥70	93 (14)	372 (12.1)	
Gender			<0.0001
Male	499 (75.3)	2,071 (67.6)	
Female	164 (24.7)	994 (32.4)	
Place of residence			0.007
Urban	487 (73.4)	2,399 (78.3)	
Rural	176 (26.6)	666 (21.7)	
Cigarette smoking [b]			<0.0001
Non-regular user	292 (44)	2,220 (72.4)	
Regular user	371 (56)	845 (27.6)	
Pack-years of cigarette smoking	20.8 ± 29.8	5.3 ± 13.1	<0.0001
Water-pipe smoking			
Non-regular user	602 (90.8)	2,858 (93.2)	0.027
Regular user	61 (9.2)	207 (6.8)	
Head-years of water-pipe smoking	48.6 ± 69.1	35.6 ± 65.5	0.083
Alcohol drinking			<0.0001
Non-user [c]	617 (93.1)	2,947 (96.2)	
Regular user	46 (6.9)	118 (3.8)	
Socioeconomic status			<0.0001
Low	400 (60.3)	1,440 (47)	
High	263 (39. 7)	1,625 (53)	
DMFT index			<0.0001
Poor	489 (73.8)	1,510 (49.3)	
Good	174 (26.2)	1,555 (50.7)	

a HNSCC: head and neck squamous cell carcinoma. Cases and controls were frequently matched on age, gender, and place of residence.
b Regular cigarette smoking: smoking a cigarette per week for at least a six-month consecutive period during the lifetime.
c Non-user included non-regular users.

Regular opium use was found to be significantly associated with an elevated risk of HNSCC. Table 11 presents the findings regarding the relationship between regular opium use and the risk of HNSCC, inclusive of all types of HNSCC combined (Table 11).

The adjusted OR for regular opium use was found to be 3.8 (95% CI: 3, 4.8). There was a robust and clear relationship between opium use and the risk of developing HNSCC, which was demonstrated by the strong dose-response association observed when evaluating factors such as the frequency of use, the quantity used, the duration of use, and the cumulative use of opium. For instance, the OR and a 95% CI was 2 (1.2, 3.5) for individuals who had consumed opium in quantities greater than the third tertile among users, in comparison to those with a cumulative use falling within the second tertile, where the OR was 2.3 (95% CI: 1.4, 3.8) (Table 11).

The two prevalent forms of opium use, Teriak (raw opium) and Shireh (opium juice), were both associated with a higher risk of HNSCC. Nevertheless, the association between Shireh and HNSCC was observed to be stronger, with an OR of 7.2 (95% CI: 4.4, 11.6), in contrast to Teriak, which had an OR of 3.4 (95% CI: 2.6, 4.4). Both methods of opium use, namely oral ingestion and smoking, were found to be

significantly associated with a higher risk of HNSCC. However, the use of opium by oral ingestion was associated with a greater risk of HNSCC, with an OR of 8.3 (95% CI: 4.7, 14.8), compared to smoking [with an OR of 2.7 (95% CI: 2.0 to 3.5)]. The strongest associations were observed in individuals who used both oral ingestion and smoking [OR: 13.0 (95% CI: 8.1, 20.6)] (Table 11).

Table 11. The associations of opium use with head and neck squamous cell carcinoma (HNSCC).

	HNSCC cases a N (%)	Controls N (%)	Adjusted OR [b] (95%CI c)	P trend [g]	P for heterogeneity
Regular Opium Use [d]					
Non-user [e]	368 (55.5)	2,664 (86.9)	Reference		
Regular user [f]	295 (44.5)	401 (13.1)	3.8 (3, 4.8)		
P for heterogeneity			<0.0001		
Duration of opium use (year)					
1st tertile (≤11)	51 (17.3)	143 (35.7)	Reference		
2nd tertile (12-23)	101 (34.2)	127 (31.6)	1.7 (1.1, 2.7)		
3rd tertile (≥24)	143 (48.5)	131 (32.7)	2.5 (1.5, 4.1)	<0.0001	<0.0001
Cumulative use [h] (gram-year)					
1st tertile (≤3.6)	38 (12.9)	134 (33.4)	Reference		
2nd tertile (3.7- 24.4)	104 (35.2)	134 (33.4)	2.3 (1.4, 3.8)		
3rd tertile (≥24.5)	153 (51.9)	133 (33.2)	2 (1.2, 3.5)	0.022	<0.0001
Frequency-Year [i]					
1st tertile (≤8)	30 (10.2)	138 (34.4)	Reference		
2nd tertile (8.1-23)	52 (17.6)	130 (32.4)	1.7 (1, 3)		
3rd tertile (≥23)	213 (72.2)	133 (33.2)	5.1 (3, 8.5)	<0.0001	<0.0001
Average intensity (gram/day)					
1st tertile (≤0.4)	62 (21.0)	150 (37.4)	Reference		
2nd tertile (0.4-2)	110 (37.3)	118 (29.4)	1.3 (0.8, 2.1)		
3rd tertile (≥2)	123 (41.7)	133 (33.2)	0.9 (0.5, 1.4)	0.460	<0.0001
Type of opium used					
Non-user	368 (55.5)	2,664 (86.9)	Reference		
Crude opium (Teriak)	238 (35.9)	360 (11.8)	3.4 (2.6, 4.4)		
Opium juice (Shireh)	57 (8.6)	41 (1.3)	7.2 (4.4, 11.6)		<0.0001
Route of opium use					
Non-user	368 (55.5)	2,664 (86.9)	Reference		
Only smoking	168 (25.3)	337 (11.0)	2.7 (2, 3.5)		
Only ingestion	35 (5.3)	28 (0.9)	8.3 (4.7, 14.8)		
Both routes	92 (13.9)	36 (1.2)	13.0 (8.1, 20.6)		<0.0001

a HNSCC: head and neck squamous cell carcinoma. Cases and controls were frequently matched on age, gender, and place of residence.
b Random-effect odds ratio. adjusted for age(categorical), gender (categorical), place of residence (categorical), pack-years of cigarette smoking (continuous), head-years of water-pipe smoking (continuous), regular alcohol drinking (categorical), socioeconomic status (categorical), and oral health (DMF index: continuous). Likelihood heterogeneity test by center.
c 95% CI:95% confidence interval.
e Non-user included non-regular users.
f Regular opium use: using opium at least once a week for at least a six-month consecutive period during the lifetime.
g P trend: P-values for trend were obtained from adjusted models by assigning values of 1, 2, and 3 to low use (Q1), moderate use (2), and high use (Q3), respectively.
h Cumulative use: Total frequency of opium use (per day) multiplied amount (gram) of opium and total duration (year).
i Frequency-Year: Total frequency of opium use (per day) multiplied by total duration (year).

Table 12. The association of opium use with head and neck squamous cell carcinoma (HNSCC) among never tobacco smokers including cigarette and water-pipe smoking.

	HNSCC case [a] N (%)	Controls N (%)	Adjusted OR [b] (95%CI [c])	P trend [g]	P for heterogeneity
Total	246	1,964			
Regular Opium Use [d]					
Non-user [e]	207 (84.2)	1,859 (94.6)	Reference		
Regular user [f]	39 (15.8)	105 (5.4)	5.2 (3.3, 8.2)		<0.0001
Duration of opium use (year)					
1st tertile (≤11)	15 (38.5)	49 (46.7)	Reference		
2nd tertile (12-23)	11 (28.2)	24 (22.9)	2.1 (0.7, 6.5)		
3rd tertile (≥24)	13 (33.3)	32 (30.5)	2.7 (0.9, 7.6)	0.058	<0.0001
Cumulative use [h] (Gram-Year)					
1st tertile (≤3.6)	11 (28.2)	41 (39)	Reference		
2nd tertile (3.7- 24.4)	17 (43.6)	38 (36.2)	2.1 (0.8, 5.6)		
3rd tertile (≥24.5)	11 (28.2)	26 (24.8)	2.4 (0.8, 7.4)	0.102	<0.0001
Frequency-Year [i]					
1st tertile (≤8)	8 (20.5)	40 (38.1)	Reference		
2nd tertile (8.1-22)	11 (28.2)	39 (37.1)	1.9 (0.6, 6)		
3rd tertile (≥23)	20 (51.3)	26 (24.8)	6.3 (2, 19.4)	0.001	<0.0001
Average intensity (gram/day)					
1st tertile (≤0.4)	17 (43.6)	50 (47.6)	Reference		
2nd tertile (0.5-2)	9 (23.1)	32 (30.5)	0.6 (0.2, 2)		
3rd tertile (≥2)	13 (33.3)	23 (21.9)	1.7 (0.5, 5.8)	0.268	<0.0001
Type of opium used					
Non-user	207 (84.2)	1,859 (94.7)	Reference		
Crude opium (Teriak)	35 (14.2)	94 (4.8)	5.1 (3.2, 8.3)		
Opium juice (Shireh)	4 (1.6)	11 (0.5)	5.8 (1.7, 19.6)		<0.0001
P for heterogeneity					
Route of opium use					
Non-user	207 (84.2)	1,859 (94.6)	Reference		
Smoking only	20 (8.1)	86 (4.4)	3.4 (1.9, 5.9)		
Oral ingestion	6 (2.4)	12 (0.6)	6.4 (2.2, 18.8)		
Both routes	13 (5.3)	7 (0.4)	24.8 (9.2, 66.9)		<0.0001

a HNSCC: head and neck squamous cell carcinoma. Cases and controls were frequently matched on age, gender, and place of residence.
b Random-effect odds ratio. adjusted for age (categorical), gender (categorical), place of residence (categorical), pack-years of cigarette smoking (continuous), head-years of water-pipe smoking (continuous), regular alcohol drinking (categorical), socioeconomic status (categorical), and oral health (DMF index: continuous). Likelihood heterogeneity test by center.
c 95% CI:95% confidence interval
d After reclassifying opium users who started opium use within 3 years prior cancer diagnosis.
e Non-user included lifetime users.
f Regular opium use: using opium at least once a week for at least a six-month consecutive period during the lifetime.
G P trend: P-values for trend were obtained from adjusted models by assigning values of 1, 2, and 3 to low use (Q1), moderate use (2), and high use (Q3), respectively

The regular use of opium was found to be associated with a significantly higher risk of developing HNSCC in the pharynx, larynx, and other sub-sites within HNSCC. The OR (95% CI) for regular opium use for these sites were 2.9 (1.4, 6.0), 6.5 (4.7, 9.0), and 5.9 (2.4, 14.7), respectively.

The study observed the association between different measurements of opium use and an increased risk of cancers of the larynx and other sub-sites within the HNSCC definition. The association between regular opium use varied among sub-sites of the larynx, with OR (95% CI) of 18.3 (8.2, 40.5) for the supraglottis, 6.2 (3.6, 10.6) for the glottis, and 4.4 (2.5, 7.7) for larynx, NOS.

In contrast, there was no significant association observed between opium use and lip and oral cavity squamous cell carcinoma, as the OR (95%CI) were 1.5 (0.9, 2.4) for regular opium use and 1.2 (0.4, 3.48) for cumulative use. Even among individuals who never smoked tobacco, the association between opium use and the risk of HNSCC remained evident (Table 12). The OR (95% CI) for the association between opium use and HNSCC in never smokers was 5.2 (3.3, 8.2).

The interaction test for those who used opium and tobacco at the same time was not different from those who used opium or tobacco, neither multiplicative interaction nor additive. The tests conducted for interactions did not yield significant results on the additive risk scale, as evidenced by the RERI (95% CI) of 3.2 (-0.5, 6.9), AP of 0.5 (-0.1, 0.9), and S of 2.2 (0.6, 8.3). This suggests that the impact of opium use could not be altered by cigarette smoking, and there was no evidence to support this notion on the multiplicative scale (P= 0.5), even when considering anatomic sub-sites (lip and oral cavity, P= 0.4; pharynx, P= 0.9; larynx, P= 0.1; other sub-sites, P= 0.2).

Despite the possibility of the participants underreporting their opium use, with a sensitivity of 0.77 in cases and 0.68 in controls, the association between HNSCC and regular opium use remained significant. The corrected OR (95% CI) was 2.5 (2.1, 3.0) (Table 13).

Table 13. The association of head and neck squamous cell carcinoma (HNSCC) and opium use based on uncorrected odds ratio OR) and OR corrected for underreporting bias.

Sites	Uncorrected OR (95%CI)	Corrected OR [a] (95%CI)
HNSCC combined	5.3 (4.4, 6.4)	2.5 (2.1, 3)
Lip and oral cavity	1 (0.7, 1.4)	0.5 (0.3, 0.8)
Pharynx	3.1 (1.7, 5.5)	1.5 (0.8, 2.8)
Larynx	16 (12.3, 20.7)	5.7 (4.5, 7.2)
Other subsites	6.6 (3.1, 14.0)	3.0 (1.4, 6.4)

[a] Corrected odds ratio for underreporting bias: 0.77 sensitivity of opium reporting in cases and 0.68 in controls (Rashidian et al. 2017)

Additionally, the surface plot analysis of the corrected OR and sensitivity of self-reported opium use, ranging from 0.5 to 0.9 in both cases and controls, demonstrated that the zone of non-significance of the corrected OR did not intersect (Figure 16).

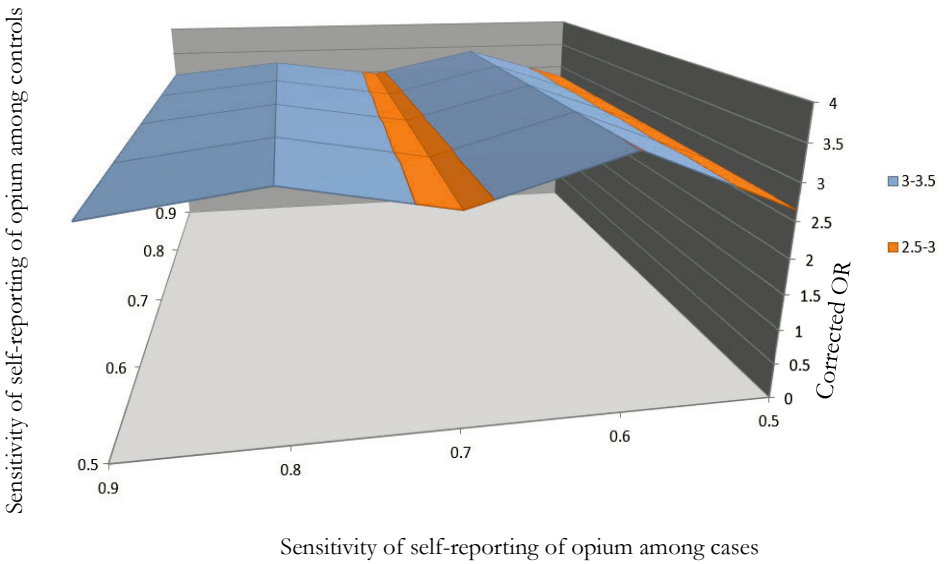

Figure 16. Surface plot of corrected odds ratio (OR) and sensitivity of self-reporting of opium in cases and controls.

5.1.3 Study III

To evaluate the association between opium use and the risk of BC, a total of 717 individuals with BC and 3,477 without BC were enrolled in the study. Among the individuals with BC who were invited to participate in the study, the non-response rate was 1%, while among controls, the non-response rate was 11%. The main reasons cited for non-response were sickness and lethargy among BC patients, and lack of time or reluctance to provide a biological sample among controls. However, there were no significant differences in gender and age distribution between those who agreed to participate and those who declined. The distribution of characteristics between cases and controls indicates that 87% of the individuals with BC were male, while 69% of the controls were male. Of the participants, 62.8% of those with BC and 62.3% of the controls were found to be residing in non-capital cities of their respective regions. The mean age of the individuals with BC was 63 years (±SD: 11.1), while the mean age of the controls was 58 years (±SD: 11.6). A significant proportion (60%) of the individuals with BC was found to be cigarette smokers, whereas only 28% of the controls reported smoking cigarettes. Additionally, 42% of the individuals with BC reported regular opium use, which was noticeably higher than the proportion of controls (13%) who reported using opium (Table 14).

Table 14. Distribution of demographic characteristics and habits of the bladder cancer (BC) cases and controls at the time of interview.

Variable Category	BC cases Number (%)	Controls Number (%)
Total	717 (100)	3,477 (100)
Age		
30-39	14 (2.0)	257 (7.4)
40-49	50 (7.0)	559 (16.1)
50-59	181 (25.2)	1070 (30.8)
60-69	267 (37.2)	1092 (31.4)
≥70	205 (28.6)	499 (14.4)
Gender		
Female	93 (13.0)	1,077 (31.0)
Male	624 (87.0)	2,400 (69.0)
Place of residence		
Capital city of the region	267 (37.2)	1310 (37.7)
Non-capital city of the region	450 (62.8)	2167 (62.3)
Province		
Tehran	139 (19.4)	816 (23.5)
Fars	166 (23.2)	943 (27.1)
Kerman	150 (20.9)	525 (15.1)
Golestan	46 (6.4)	374 (10.8)
Mazandaran	24 (3.4)	136 (3.9)
Kermanshah	52 (7.3)	251(7.2)
Khorasan-Razavi	30 (4.2)	170 (4.9)
Bushehr	56 (7.8)	84 (2.4)
Hormozgan	27 (3.8)	78 (2.2)
Systan-Balouchestan	27 (3.8)	100 (2.9)
Cigarette smoking		
Non-smoker	287 (40.0)	2,500 (71.9)
Smoker [a]	430 (60.0)	977 (28.1)
Cigarette pack-years		
Non-smoker	287 (41.4)	2,500 (71.9)
Light <15)	111 (15.5)	449 (12.9)
Moderate (15-31)	120 (16.7)	255 (7.3)
Heavy (>31)	184 (25.7)	229 (6.6)
Unknown	15 (2.1)	44 (1.3)
Occupation		
High-skilled white collar	202 (28.2)	1,011 (29.1)
Low-skilled white collar	153 (21.3)	575 (16.5)
High-skilled blue collar	273 (38.1)	966 (27.8)
Low-skilled blue collar	89 (12.4)	925 (26.6)
Opium use		
Non-user [b]	389 (54.3)	2,887 (83.02)
Irregular	27 (3.8)	139 (4.0)
Regular [c]	301 (42.0)	451 (13.0)
Type of opium used		
Crude opium (Teriak)	249 (34.7)	397 (11.4)
Opium juice (Shireh)	20 (2.8)	30 (0.9)
Both types	32 (4.5)	24 (0.7)
Route of opium use		
Only smoking	207 (28.9)	376 (10.8)

Table 14 (continued)

Variable Category	BC cases	Controls
Only ingestion	27 (3.8)	28 (0.8)
Both routes	65 (9.1)	45 (1.3)
Unknown	2 (0.1)	2 (0.1)
Cumulative use of opium (kg)d		
Non-user	389 (54.3)	2,887 (83.0)
<4	95 (13.2)	218 (6.3)
4 -16	92 (12.8)	117 (3.4)
>16	111 (15.5)	114 (3.3)
Duration of opium use (years)		
<17	76 (10.6)	212 (6.1)
17 -28	106 (14.8)	141 (4.1)
>28	119 (16.6)	98 (2.8)
Daily dose of opium (gram)		
<1	135 (18.8)	236 (6.8)
1-2	52 (7.3)	94 (2.7)
>2	114 (15.9)	121 (3.5)
Count per day (frequency)e		
<1	126 (17.6)	314 (9.0)
1-2	116 (16.2)	101 (2.9)
>2	59 (8.2)	36 (1.0)
Average amount of opium used each time (gram) f		
<1	149 (20.8)	199 (5.7)
1-2	63 (8.8)	142 (4.1)
>2	89 (12.4)	110 (3.2)
Starting age of opium use		
<20	42 (5.9)	88 (2.5)
20-29.99	68 (9.5)	106 (3.1)
30-39.99	106 (14.8)	112 (3.2)
≥40	96 (13.4)	162 (4.7)
Time since stopping opium use		
Current user	198 (27.6)	239 (6.9)
<10 years	76 (10.6)	131 (3.8)
≥10 years	27 (3.8)	81 (2.3)

a Smoking a cigarette per week for at least a six-month consecutive period during the lifetime.
b Those who started opium use < 3 years before the index date considered as non-users.
c Using opium at least once a week for at least a six-month consecutive period during the lifetime.
d Cumulative use: total duration of opium use (days) multiplied average daily amount (grams) of opium.
e Weighted average of daily frequency of opium use.
f Weighted average of the amount of opium used each time.

The OR of developing BC among individuals who reported regular opium use was 3.5 compared to those who did not use opium (95% CI: 2.8, 4.3) (Table 15). As a side finding, it was observed that heavy cigarette smokers had a 3.7-fold risk of developing BC compared to individuals who did not smoke cigarettes, as was expected based on what is known about the risk of BC and smoking (Mori et al. 2020; Rink et al. 2015).

Table 15. Characteristics of opium use among regular opium users, and the odds ratios (OR) for opium use with bladder cancer from a model including age, gender, province, cigarette pack-years and opium use.

Variable Category	OR (95% CI [a])
Cigarette pack-years	
Non-smoker	Ref.
Light (<15)	1.5 (1.2, 2.0)
Moderate (15-31)	2.5 (1.9, 3.3)
Heavy (>31)	3.7 (2.8, 4.8)
Opium use [c]	
Non-user	Ref.
Irregular	1.1 (0.7, 1.7)
Regular [b]	3.5 (2.8, 4.3)

[a] 95% confidence interval.
[b] Regular opium use: using opium at least once a week for at least a six-month consecutive period during the lifetime.

The other metrics of opium use, such as type of opium, route of administration, lifelong use, duration, daily dose, frequency, average use, starting age, and time since cessation of opium use, were strongly associated with the development of BC among regular opium users. After adjusting for other factors, we found that individuals who had used both teriak and shireh had a 7.3-fold risk of developing BC compared to non-users (OR: 7.3, 95% CI: 4.1, 13.1). Both routes of opium use, i.e., smoking and ingestion, were strongly associated with a higher risk of BC. Additionally, individuals who used both routes of opium at the same time had an even higher risk of developing BC (OR:6.9, 95% CI: 4.4, 10.7). Lifelong regular use of opium was associated with an increasing risk of developing BC, compared to non-users. Individuals who had used less than 4 kg of opium during their lifetime had a 2-fold higher risk of BC (OR: 2.3, 95% CI: 1.7, 3.2). The risk of BC was significantly higher among those who had used between 4 to 16 kg of opium during their lifetime, compared to non-users (OR: 4.3, 95% CI: 3.1, 6.1). Furthermore, individuals who had used more than 16 kg of opium during their lifetime had an even higher risk of developing BC (OR: 4.9, 95% CI: 3.5, 6.9) (Table 16).

The duration of regular opium use was strongly associated with a higher risk of BC. Individuals who had used opium regularly for less than 17 years had a 2-fold risk of BC (OR: 2.2, 95% CI: 1.6, 3.1). The risk of BC increased almost 5-fold with an increase in duration between 17 and 28 years, and more than 28 years (OR: 4.5, 95% CI: 3.2, 6.1), (OR: 4.6, 95% CI: 3.3, 6.4) respectively. In addition, the frequency of regular opium use was strongly associated with an increasing risk of BC. Opium users with a frequency of less than one time per day had a higher risk of BC (OR: 2.1, 95% CI: 1.6, 2.8). The risk of BC continued to increase with a frequency of one and two times per day, and more than two times per day, respectively. Moreover, the average amount of opium used at a given time was found to be positively associated with the risk of BC (p-trend=0.0001). Additionally, the time since stopping the use of opium was also found to have an impact on the risk of BC. Current opium users had a higher risk of BC with an OR of 3.8 (95%

CI: 3.0, 4.9). However, the risk of BC decreased as the time since stopping opium use increased, with an OR of 2.7 (95% CI: 2.7, 3.9) (Table 16).

Table 16. Odds ratios (OR) for opium use with bladder cancer, by metrics of opium use among regular opium users adjusted for age, gender, province, cigarette pack-years.

Metric of opium use Category	OR (95%CI)
Type of opium used	
Crude opium (Teriak)	3.2 (2.5, 4.1)
Opium juice (Shireh)	4.0 (2.2, 7.4)
Both types	7.3 (4.1, 13.1)
Route of opium use	
Only smoking	2.8 (2.2, 3.6)
Only ingestion	6.7 (3.7, 12.1)
Both routes	6.9 (4.4, 10.7)
Cumulative use of opium (kg)[a]	
Non-user	
<4	2.3 (1.7, 3.2)
4 -16	4.3 (3.1, 6.1)
>16	4.9 (3.5, 6.9)
Duration of opium use (years)	
<17	2.2 (1.6, 3.1)
17 -28	4.5 (3.2, 6.1)
>28	4.6 (3.3, 6.4)
Daily dose of opium (g)	
<1	2.9 (2.2, 3.8)
1-2	3.1 (2.1, 4.6)
>2	4.9 (3.6, 6.8)
Count per day (frequency)[b]	
<1	2.1 (1.6, 2.8)
1-2	6.5 (4.6, 9.1)
>2	9.6 (6.0, 15.5)
Average of opium use at a time (g) [c]	
<1	3.9 (2.9, 5.1)
1-2	2.4 (1.7, 3.5)
>2	4.1 (2.95.8)
	p trend 0.0001
Starting age of opium use	
<20	2.4 (1.4, 4.2)
20-29.99	3.2 (2.3, 4.4)
30-39.99	4.8 (3.5, 6.6)
≥40	2.5 (1.9, 3.4)
Time since stopping opium use	
Current	3.8 (3.0, 4.9)
<10	2.7 (2.0, 3.9)
≥10	1.3 (0.8, 2.2)

[a] Cumulative use: total duration of opium use (days) multiplied average daily amount (grams) of opium.
[b] Weighted average of daily frequency of opium use
[c] Weighted average of opium use at each time

The study found evidence of an interaction of tobacco smoking on the association between opium use and the risk of BC. The REIRI was 2.6 (95% CI: (0.8, 4.4), which means that the joint effect of opium use and tobacco use on the risk of BC is greater than the sum of their individual effects. The 95% CI did not include 0, indicating that the interaction was statistically significant. Additionally, the AP was 0.35 (95% CI: 0.1, 0.6), meaning that 35% of the cases of BC among opium users can be attributed to the interaction between opium use and tobacco use. The S was 1.7 (95% CI: 1.1, 2.4), which also suggests a synergistic effect between the two risk factors. However, the study also found that among non-tobacco users, the association between opium use and the risk of BC was still strong, suggesting that the effect of opium use on BC risk is not limited to those who smoke cigarettes. Table 17 provides information on the association between BC and opium use, tobacco use, and the combined use of both substances. The results show a higher risk of BC for those who use both tobacco and opium compared to those who use only one or neither substance.

Table 17. Odds ratios (OR) and 95% confidence interval (CI) of regular opium use and tobacco interaction for bladder cancer (BC) adjusted for age and gender.

	Cigarette non-smoker	Cigarette smoker
	OR (95% CI)	
Non-user of opium	Ref.	2.4 (2.0, 3.0)
Opium user *	3.5 (2.3, 5.1)	7.5 (5.9, 9.5)

*Results for irregular opium use not reported

The strong association between opium use and BC remained even after controlling for the effect of tobacco smoking. Additionally, the various metrics of opium use, such as type of opium, route of use, cumulative amount, duration, daily dose, frequency, and the average amount of opium in grams used at a given time, were all strongly associated with an increased risk of BC among those who did not use tobacco regularly. The interesting finding regarding the starting age of opium use and its association with the risk of BC shows that the risk of BC is highest among those who started using opium at a younger age (between 20 to 20.99 years old) (OR: 6.7, 95%CI: 3.1, 14.6) and decreases as the starting age of users increases (over 40 years old) (OR: 2.1, 95%CI: 1.2, 3.7). On the other hand, the risk of developing BC is higher the longer it has been since the person has stopped using opium. Current opium users are at a four times greater risk (OR: 4.3, 95% CI: 2.8, 6.5), while those who have not used opium for over 10 years have a three times greater risk (OR: 2.6, 95% CI: 0.5,13.1).

The quantitative bias analysis to assess non-differential bias by considering the sensitivity of self-reporting (the sensitivity among cases was 0.77 and among controls 69%) showed that, despite this potential bias, the study still found a significant association between regular opium use and BC (OR: 5.6, 95% CI: 3.9, 9.1) (Table 18).

Table 18. Odds ratios (OR) and 95% confidence interval (CI) of bladder cancer for opium observed and corrected for non-differential bias among regular opium users.

	Opium*		Total
	Use	Non-use	
Bladder cancer cases	301	389	690
Controls	451	2,887	3,338
Observed OR, 95% CI	4.9 (4.1, 5.9)		
Corrected OR, 95% CI	5.6 (3.9, 9.1)		

Specificity for cases and controls 100%, sensitivity among cases 70% and among controls 69%.
*Irregular opium users were excluded

5.1.4 Study IV

This study evaluating the association between CRC and opium use included 455 instances of colon cancer and 393 instances of rectal cancer; 482 (56.8%) of the cases and 2,205 (68.6%) of the controls were male (Table 19).

Among the 848 cases, 89 individuals (10.5%) were regular opium users, while among the 3,215 controls, 439 individuals (13.7%) reported regular opium use (Table 19). It was found that there was a 30% higher risk of CRC in individuals who irregularly used opium, but this difference was not statistically significant. On the other hand, there was no significant association between regular opium use and the risk of CRC overall (OR: 0.9, 95% CI: 0.7, 1.2) or in any of its two subsites (Table 19).

Table 19. Distribution of demographic characteristics and life habits of colorectal cancer (CRC) cases and controls at the time of interview.

Variable Category	CRC cases Number (%)	Controls Number (%)
Total	848 (93.0)	3,215 (100)
Age		
30-39	66 (7.8)	250 (7.8)
40-49	123 (14.5)	505 (15.7)
50-59	235 (27.7)	997 (31.0)
60-69	252 (29.7)	1,020 (31.7)
≥70	172 (20.3)	443 (13.8)
Gender		
Female	366 (43.2)	1,010 (31.4)
Male	482 (56.8)	2,205 (68.6)
Place of residence		
Capital city of the region	538 (63.4)	1,254 (39.0)
Other	310 (36.6)	1,961 (61.0)
Province		
Tehran	170 (20.0)	816 (25.4)
Fars	248 (29.3)	943 (29.3)
Kerman	104 (12.3)	525 (16.3)
Golestan	140 (16.5)	374 (11.6)
Mazandaran	59 (7.0)	136 (4.2)
Kermanshah	66 (7.8)	251(7.8)
Khorasan-Razavi	61 (7.2)	170 (5.3)
SES		
Low	325 (38.3)	863 (26.8)
Moderate	230 (27.1)	1,085 (33.8)
High	293 (34.6)	1,267 (39.4)
Body shape at age 15		
Normal	662 (78.1)	2,694 (83.8)
Overweight	99 (11.7)	303 (9.4)
Obese	87 (10.3)	218 (6.8)
Physical activity at work		
Sedentary	282 (33.3)	1,036 (32.2)
Medium	151 (17.8)	701 (21.8)
High	176 (20.8)	695 (21.6)
Unknown	239 (28.2)	783 (24.4)

As a side finding, it was observed that red meat intake had a significant association with the risk of CRC, which showed a higher risk for colon than rectum cancer as was expected on the basis of what is known the risk of CRC and red meat consumption (Ubago-Guisado et al. 2021). Moreover, individuals with obesity have a 30% higher risk of developing colorectal cancer (CRC) compared to individuals with a normal body shape as this has been reported previously (Liu et al. 2019). In addition, individuals who have first-degree relatives with HNPCC have a 70% higher risk of developing CRC compared to individuals without such a family history, with similar ORs for cancers of the colon and rectum in parallel with other studies (Peltomäki, Olkinuora, and Nieminen 2020; M. L. Slattery et al. 2003; Martha L. Slattery and Kerber 1994). Furthermore, those who have been divorced or widowed have a higher risk of developing CRC compared to individuals who are single or married (Feng et al. 2018) (Table 20).

Table 20. Odds ratios (OR) and 95% confidence intervals (CI), adjusted for age, gender, province, and all other variables listed in this table.

Variable Category	Controls N (%)	Colorectal cancer		Colon cancer		Rectal cancer	
		N (%)	OR (95% CI)	N (%)	OR (95% CI)	N (%)	OR (95% CI)
Total	3,215 (100)	848 (93.0)		455 (49.8)		393 (43.0)	
Opium use							
Non-user	2,649 (82.4)	719 (84.8)	Ref.	386 (84.8)	Ref.	333 (84.7)	Ref.
Irregular	127 (3.9)	40 (4.7)	1.3 (0.9, 1.9)	25 (5.5)	1.4 (0.9, 2.3)	15 (3.8)	1.1 (0.6, 2.0)
Regular [a]	439 (13.7)	89 (10.5)	0.9 (0.7, 1.2)	44 (10)	0.8 (0.6, 1.2)	45 (11.5)	1.0 (0.7, 1.4)
Cofactors included in the model							
Marital Status							
Married	2,913 (90.6)	701 (82.7)	Ref.	374 (82.2)	Ref.	327 (83.2)	Ref.
Widow	164 (5.1)	99 (11.7)	1.9 (1.4, 2.6)	56 (12.3)	2.1 (1.4, 3.0)	43 (10.9)	1.8 (1.2, 2.6)
Divorced/separated	39 (1.2)	21 (2.5)	2.0 (1.1, 3.4)	10 (2.2)	1.8 (0.9, 3.9)	11 (2.8)	2.2 (1.0, 4.4)
Single	90 (2.8)	21 (2.5)	0.8 (0.5, 1.4)	12 (2.6)	1.0 (0.5, 1.8)	9 (2.3)	0.7 (0.4, 1.6)
Unknown	9 (0.3)	6 (0.7)	-	3 (0.7)	-	3 (0.8)	-
SES							
Low	863 (26.8)	325 (38.3)	Ref.	173 (38)	Ref.	152 (38.7)	Ref.
Moderate	1,085 (33.8)	230 (27.1)	0.7 (0.5, 0.8)	124 (27.3)	0.7 (0.5, 0.9)	106 (27.0)	0.6 (0.5, 0.8)
High	1,267 (39.4)	293 (34.6)	0.7 (0.6, 0.9)	158 (34.7)	0.8 (0.6, 1.0)	135 (34.4)	0.7 (0.5, 0.9)
Body shape							
Normal	2,694 (83.8)	662 (78.1)	Ref.	346 (76)	Ref.	316 (80.4)	Ref.
Overweight	303 (9.4)	99 (11.7)	1.1 (0.9, 1.5)	60 (13.2)	1.3 (0.9, 1.8)	39 (9.9)	1.0 (0.7, 1.4)
Obese	218 (6.8)	87 (10.3)	1.3 (1.0, 1.8)	49 (10.8)	1.4 (1.0, 2.0)	38 (9.7)	1.3 (0.9, 1.9)
Perceived physical workload							
Sedentary	1,036 (32.2)	282 (33.3)	Ref.	157 (34.5)	Ref.	125 (31.8)	Ref.
Medium	701 (21.8)	151 (17.8)	0.8 (0.6, 0.9)	78 (17.1)	0.7 (0.5, 1.0)	73 (18.6)	0.8 (0.6, 1.1)
High	695 (21.6)	176 (20.8)	0.8 (0.6, 1.0)	93 (20.4)	0.8 (0.6, 1.0)	83 (21.1)	0.8 (0.6, 1.1)
Unknown	695 (21.6)	239 (28.2)	0.6 (0.5, 0.8)	127 (27.9)	0.6 (0.4, 0.9)	112 (28.5)	0.6 (0.4, 0.9)
Family history of cancer							
No	3,060 (95.2)	781 (92.1)	Ref.	419 (92.1)	Ref.	362 (92.1)	Ref.
Yes	155 (4.8)	67 (7.9)	1.7 (1.3, 2.3)	36 (7.9)	1.7 (1.2, 2.6)	31 (7.9)	1.7 (1.1, 2.6)

Red meat (g/day)							
< 12.82	1,742 (54.2)	410 (48.4)	Ref.	215 (47.3)	Ref.	195 (49.6)	Ref.
12.83-25.64	990 (30.8)	233 (27.5)	1.3 (1.0, 1.5)	119 (26.2)	1.3 (1.0, 1.6)	114 (29.0)	1.3 (1.0, 1.7)
>25.64	480 (14.9)	205 (24.2)	2.2 (1.8, 2.7)	121 (26.6)	2.5 (1.9, 3.3)	84 (21.4)	1.9 (1.4, 2.6)
Unknown	3 (0.01)	-		-	-	-	-
Fruit and vegetables (g/day)							
<422	1,606 (50.0)	409 (48.2)	Ref.	211 (46.4)	Ref.	198 (50.4)	Ref.
422-576	803 (25.0)	200 (23.6)	1.0 (0.8, 1.3)	108 (23.7)	1.2 (0.9, 1.5)	92 (23.4)	0.9 (0.7, 1.2)
>576	803 (25.0)	239 (28.2)	1.1 (0.9, 1.3)	136 (29.9)	1.3 (1.0, 1.7)	103 (26.2)	0.9 (0.7, 1.2)
Unknown	3 (0.01)	-		-	-	-	-

[a] Using opium at least once a week for at least a six-month consecutive period during the lifetime

In analyses focusing on the characteristics of opium use, the ingestion of opium was found to be associated with an increased risk of developing rectal cancer, but not colon cancer. Specifically, the OR for the association between opium ingestion and rectal cancer was 2.3 (95% CI: 1.0, 5.6). However, the average amount of opium used each time was not found to be associated with an increased risk of CRC (Table 21).

The risk of developing CRC was found to increase as the frequency of opium use per day increased. Individuals who used opium more than two times per day were found to have 2.0 times higher OR (95% CI: 1.1, 3.8) for CRC compared to individuals who did not use opium. The OR for colon cancer was determined to be 2.0 (95% CI: 1.0, 4.5), while the OR for rectal cancer was also 2.0 (95% CI 0.9, 4.4). The quadratic p-trend related to the frequency of opium use and risk of CRC was found to be 0.008 for CRC, 0.02 for colon cancer, and 0.1 for rectal cancer (Table 21).

Table 21. Odds ratios (OR) and 95% confidence intervals (CI) by the characteristics of regular opium use from models adjusted for age, gender, province, marital status, family history of cancer, red meat, vegetables, body shape, socio-economic status, perceived physical workload. Lag 3 years.

Metric of opium use Category	Controls	Colorectal cases		Colon cases		Rectal cases	
	N (%)	N (%)	OR (95% CI)	N (%)	OR (95% CI)	N (%)	OR (95%CI)
Type of opium used							
No opium use	2,649 (82.4)	719 (84.8)	Ref.	386 (84.8)	Ref.	333 (84.7)	Ref.
Crude opium (Teriak)	387 (12.0)	72 (8.5)	0.8 (0.6, 1.1)	34 (7.5)	0.8 (0.5, 1.2)	35 (8.9)	0.9 (0.6, 1.3)
Opium juice (Shireh)	31 (1.0)	9 (1.1)	1.3 (0.6, 2.8)	5 (1.1)	0.9 (0.3, 2.8)	7 (1.2)	1.6 (0.6, 4.4)
Both types	21 (0.7)	8 (0.9)	1.6 (0.7, 3.6)	5 (1.1)	1.6 (0.5, 4.9)	3 (0.8)	1.5 (0.5, 4.5)
Route of opium use							
Only smoking	368 (11.5)	69 (8.1)	0.9 (0.6, 1.2)	34 (7.5)	0.8 (0.6, 1.2)	35 (8.9)	0.9 (0.6, 1.3)
Only ingestion	27 (0.8)	12 (1.4)	1.6 (0.8, 3.2)	5 (1.1)	1.0 (0.4, 2.8)	7 (1.8)	2.3 (1.0, 5.6)
Both routes	41 (1.3)	8 (0.9)	0.9 (0.4, 2.0)	5 (1.1)	1.0 (0.4, 2.6)	3 (0.8)	0.8 (0.2, 2.7)
Unknown	3 (0.1)	-	-	-	-	-	-
Count per day [a]							
< 1	308 (9.6)	48 (5.7)	0.7 (0.5, 1.0)	23 (5.1)	0.6 (0.4, 1.0)	25 (6.4)	0.8 (0.5, 1.2)
1-2	97 (3.0)	24 (2.8)	1.1 (0.7, 1.8)	12 (2.6)	1.1 (0.6, 2.1)	12 (3.0)	1.2 (0.6, 2.3)
> 2	34 (1.1)	17 (2.0)	2.0 (1.1, 3.8)	9 (2)	2.0 (1.0, 4.5)	8 (2.0)	2.0 (0.9, 4.4)
Average opium uses at a time (g)							
< 1	190 (5.9)	54 (6.4)	1.2 (0.9, 1.9)	28 (6.2)	1.1 (0.7, 1.8)	26 (6.6)	1.4 (0.9, 2.1)
1-2	140 (4.4)	18 (2.1)	0.6 (0.4, 1.0)	7 (1.5)	0.4 (0.2, 1.0)	11 (2.8)	0.7 (0.4, 1.4)
> 2	109 (3.4)	17 (2.0)	0.7 (0.4, 1.2)	9 (2)	0.8 (0.4, 1.6)	8 (2.0)	0.7 (0.3, 1.4)
Daily dose of opium (g) [b]							
< 1	233 (7.3)	46 (5.4)	0.9 (0.6, 1.2)	23 (5.1)	0.8 (0.8, 1.3)	23 (5.9)	1.0 (0.6, 1.6)
1-2	84 (2.6)	17 (2.0)	0.9 (0.5, 1.6)	9 (2)	0.9 (0.4, 1.9)	8 (2.0)	0.9 (0.4, 1.8)
> 2	122 (3.8)	26 (3.1)	1.0 (0.6, 1.5)	12 (2.6)	0.9 (0.5, 1.8)	14 (3.6)	1.1 (0.6, 1.9)
Starting age of opium use							
< 20	51 (1.6)	9 (1.1)	0.8 (0.4, 1.8)	5 (1.1)	0.9 (0.3, 2.3)	4 (1.0)	0.8 (0.3, 2.3)
20-29	134 (4.2)	20 (2.4)	0.7 (0.4, 1.2)	9 (2.0)	0.6 (0.3, 1.2)	11 (2.8)	0.9 (0.5, 1.7)
30-39	106 (3.3)	29 (3.4)	1.3 (0.8, 2.0)	16 (3.5)	1.3 (0.7, 2.3)	13 (3.3)	1.2 (0.7, 2.2)
≥ 40	148 (4.6)	31 (3.7)	0.8 (0.6, 1.3)	14 (3.1)	0.7 (0.4, 1.3)	17 (4.3)	1.0 (0.6, 1.7)
Time since stopping opium use							
Current user	213 (6.6)	41 (4.8)	0.9 (0.6, 1.3)	25 (5.5)	1.1 (0.9, 2.3)	16 (4.1)	0.8 (0.4, 1.3)

Table 21 (continued)

Metric of opium use Category	Controls	Colorectal cases		Colon cases		Rectal cases	
	N (%)	N (%)	OR (95% CI)	N (%)	OR (95% CI)	N (%)	OR (95%CI)
Unknown	7 (0.2)	1 (0.1)	-	-	-	1 (0.3)	-
Duration of opium use (years)							
< 19	254 (7.9)	60 (7.1)	1.0 (0.8, 1.4)	26 (5.7)	0.9 (0.6, 1.3)	34 (8.7)	1.2 (0.8, 1.8)
19-29	107 (3.3)	19 (2.2)	0.9 (0.5, 1.5)	13 (2.9)	1.2 (0.6, 2.2)	6 (1.5)	0.6 (0.3, 1.4)
> 29	78 (2.4)	1 (1.2)	0.5 (0.3, 1.1)	5 (1.1)	0.5 (0.2, 1.2)	5 (1.3)	0.6 (0.2, 1.5)
Cumulative amount of opium (kg) [c]							
< 4	211 (6.6)	45 (5.3)	0.9 (0.7, 1.3)	22 (2.2)	0.8 (0.5, 1.3)	23 (5.9)	1.1 (0.7, 1.7)
4 -14	115 (3.6)	24 (2.8)	1.0 (0.6, 1.5)	10 (2.2)	0.8 (0.4, 1.5)	14 (3.6)	1.2 (0.6, 2.1)
>14	112 (3.5)	18 (2.1)	0.7 (0.4, 1.3)	11 (2.4)	1.0 (0.5, 1.7)	7 (1.8)	0.6 (0.3, 1.3)
Unknown	1 (0.0)	2 (0.2)	-	1(0.2)	-	1 (0.3)	-
Cumulative count of opium uses (times) [d]							
<4,900	217 (6.8)	35 (4.1)	0.7 (0.5, 1.0)	17 (3.7)	0.7 (0.4, 1.1)	18 (4.6)	0.8 (0.5, 1.3)
4,900-11,000	126 (3.9)	30 (3.5)	1.1 (0.7, 1.8)	15 (3.3)	1.1 (0.6, 1.9)	15 (3.8)	1.2 (0.7, 2.2)
>11,000	96 (3)	24 (2.8)	1.1 (0.7, 1.7)	12 (2.6)	1.0 (0.5, 1.8)	12 (3.1)	1.2 (0.6, 2.2)

[a] Duration-weighted average of the period-specific daily frequencies of opium use
[b] Count per day multiplied by the average of opium use at a time (g)
[c] Cumulative amount: the average daily amount of opium multiplied by the total duration of opium use (days)
[d] Cumulative count: average count per day multiplied by the total duration of opium use (days)

6 DISCUSSION OF MAJOR FINDINGS

First, the validation and pilot phases yielded results regarding the feasibility of the main study. Our team was able to successfully design, validate, and check the reliability of the questionnaires used in the study. Furthermore, the response rate among both cases and controls was high. We were able to match the controls for age, gender, province, and place of residence, but perfect matching was not achievable due to the younger controls and a higher proportion of controls from the Tehran and Fars centers, the two active regional centers that became involved in the study at an early stage.

A significant variation was observed in the prevalence of opium use among participants from the ten provinces, with the highest rates being found in the eastern regions (namely Kerman, Sistan-Baluchestan, and Khorasan-Razavi). Across all provinces, the cancer patients reported higher levels of opium use compared to the control group. Specifically, the IROPICAN study recruited 3,247 cancer patients, including 914 with CRC, 717 with BC, and 918 with HNSCC, as well as 3,477 control participants. Undoubtedly, the IROPICAN study has already made significant contributions to our understanding of the potential carcinogenicity of opium use (Bidary et al., 2021; IARC Monographs Vol 126 group, 2020; M Filho et al., 2023; Mansouri et al., 2022; Mohebbi, et al., 2021; Singh et al., 2021).

The use of opium was associated with a significantly higher risk of BC, HNSCC, as well as specific anatomic subsites of HNSCC, such as cancers of the pharynx, larynx, and other subsite groups in participants with opium use compared to those who have never used opium. However, opium use was not found to be associated with the risk of CRC.

To assess the association between regular opium consumption and cancer, we found that regular opium use was associated with a roughly 4-fold increase in the risk of HNSCC and BC compared to individuals who had never used opium. This increased risk was consistent across individuals with different subsites of HNSCC such as pharynx, larynx and confirmed urothelial histology of BC and those BC cases with other or unknown histology, as well as across both males and females. However, regular opium use does not appear to increase the risk of cancers of the CRC and HNSCC subsites, including the lips and oral cavity.

Furthermore, it was found that individuals who used both crude opium and opium juice had a 7-fold increase in the risk of HNSCC and BC. Ingesting opium was associated with a higher risk of HNSCC and BC compared to smoking it. The risk of HNSCC and BC also increased with a longer duration of opium use, with those using opium for more than 17 years having a higher risk, as well as with greater cumulative use, with those using more than 4 kg of opium having a higher risk in comparison with never opium users. Also, the risk of HNSCC and BC increased as the frequency of daily opium use increased, but the average amount of opium used each time did not have a significant effect on the risk of both HNSCC and BC. The age at which opium use began did not independently contribute to the risk of BC.

The finding is consistent with the results of five prior case-control studies regarding opium use and the risk of cancer (Alizadeh et al. 2020; Bakhshaee et al. 2017; Khoo 1981; Mousavi et al. 2003; Shoffel-

Havakuk et al. 2018). The overall consistency of the association found in multiple studies supports the idea of a causal relationship between opium use and the development of HNSCC. The increased risk of HNSCC associated with opium use may vary depending on the specific subsite within the head and neck region. The association between opium use and HNSCC was particularly strong for laryngeal cancer, which is consistent with the findings of previous literature studies (Alizadeh et al. 2020; Bakhshaee et al. 2017; Bidary et al. 2021; Khoo 1981; Mansouri et al. 2022; Mousavi et al. 2003; Singh et al. 2021).

A few studies have evaluated the association between opium use and BC risk with a limited sample size in case-control studies (Afshari et al., 2017; Akbari et al., 2015; Ghadimi et al., 2015; Sheikh, et al., 2020; Warnakulasuriya et al., 2020). According to the findings, the risk of developing BC is three times higher in individuals who consume opium compared to those who do not use opium (OR: 3.4, 95% CI: 2.7, 4.3), which is consistent with the results of earlier case-control studies that investigated the association between opium consumption and BC risk. One study reported an OR of 5.0 (95% CI: 1.1, 2.3) indicating a positive association between opium use and an increased risk of BC (Ghadimi et al. 2015), and another one showed an OR of 3.9 (95% CI: 1.2, 12) (Akbari et al. 2015). In addition, a systematic review demonstrated the association between opium consumption and a higher risk of BC in comparison to non-users, with a combined OR of 3.9 (95% CI: 3.1, 4.9) (Afshari et al. 2017). Another systematic review that investigated opium as a carcinogen, reported a fixed effect model analysis pooled OR of 4.1 (95% CI: 3.2, 5.1) and a random effect model analysis pooled OR of 3.8 (95% CI: 2.7, 5.4) (Bidary et al. 2021). Also, the results of a recent systematic review showed the overall meta-relative risk for ever or regular opium users versus never users for BC to be 4.1 (mRR: 4.1, 95% CI: 3.2, 5.1) (Filho et al. 2023). A case-control study conducted in the Kerman province of Iran, which investigated the association between opium consumption and the risk of BC in patients diagnosed from 2013 to 2015 (slightly earlier than the cases of our study, which was conducted from 2016 to 2020) compared with neighborhood controls, found that regular opium use was associated with the risk of BC compared to non-opium users with the OR of 4.4 (95% CI: 2.9, 6.5). This finding is consistent with the OR observed in this study, despite differences in the selection of controls and methods of analysis (Rashidian et al. 2021). This study also specifically looked at urothelial BC, and the findings are consistent with the results of an other study which demonstrated that individuals with a history of opium use were more likely to develop urothelial BC than those without such a history (OR: 3.0, 95% CI: 1.6, 5.4) (Zeighami et al. 2018).

The study related to opium use and the risk of CRC found that individuals who consumed opium at least twice a day had roughly a two-fold increased risk of CRC compared to those who had never used opium. Neither the amount of opium consumed during each use nor the duration or frequency of opium use appeared to have an impact on the risk of CRC. The findings related to the type and method of opium use were also not particularly strong.

There have been only a few studies conducted on the association between opium use and the risk of CRC, including two small case-control studies (Khosravizadegan et al., 2017; Naghibzadeh-Tahami et al., 2016) and one cohort study (Sheikh, et al., 2020). The present study did not find significant association between regular opium use and the risk of CRC, which is consistent with the results of a cohort study conducted by Sheikh et al. (Sheikh, et al., 2020). By contrast, two case-control studies reported a four-fold increased risk of CRC among individuals who used opium (Khosravizadegan et al. 2017; Naghibzadeh-Tahami et al. 2016). Both of these studies used neighborhood controls. There is a concern that using neighborhood controls may lead to underestimating the use of opium since opium is an illegal substance, and users may not report their use accurately (Mohebbi, et al., 2021). In this study, the controls were hospital visitors who

were interviewed in the same manner as the cases. It has been demonstrated that in such a setting, the reporting of opium use is equally accurate among both the cases and controls (Rashidian et al. 2017).

The exact mechanism by which opium causes cancer is not fully understood. The working group of the IARC discovered compelling evidence indicating that opium dross and opium pyrolysates possess properties that are typically associated with cancer-causing agents or carcinogens (IARC Monographs Vol 126 group 2020; Warnakulasuriya et al. 2020). Another explanation is that the consumption of opium may promote the development of tumors by affecting processes such as angiogenesis (the formation of new blood vessels), immunosuppression (the suppression of the immune system's response), and the promotion of cancer cell growth and division (Grandhi et al., 2017; Sheikh, et al., 2020; Vallejo et al., 2004). In addition, the likelihood of bladder exposure to carcinogens may increase as a result of opium use, since the alkaloids found in opium can cause urinary retention and cystitis, which can lead to prolonged exposure of the bladder to potentially harmful substances (Kamangar et al. 2014). However, the potential mechanisms underlying the observed variations in the association between opium use and HNSCC across different anatomic subsites are unclear.

This study indicates that smoking opium was associated with a lower risk of HNSCC and BC compared to consuming opium by ingestion. Furthermore, even after taking into account the length of time opium was used, the ingestion route still demonstrated a higher risk of HNSCC and BC. This is consistent with the findings of a study by Sheikh et al., which reported the OR of 2.6 (95% CI: 1.2, 5.4) for opium smoking and 3.8 (95% CI: 1.6, 8.9) for opium ingestion route (Sheikh, et al., 2020).

A new and noteworthy finding of this study was that individuals who had stopped using opium more than 10 years prior to the index date demonstrated a significantly reduced risk of BC compared to those who had used an equivalent amount of opium but had not stopped. This finding is unique and, to the best of our knowledge, has not been previously reported in any other study.

Furthermore, to the best of our knowledge, this study is the first to assess the impact of the frequency of opium use on the risk of CRC. In this dataset, a trend was observed between the risk of CRC and an increasing daily frequency of opium use, although this trend was not statistically significant. Furthermore, using opium at least two times per day was associated with an OR of 2.0 for both colon cancer and rectal cancer. The increased risk appeared to be particularly concentrated among individuals who used the ingestion route for opium consumption. If these findings are not due to chance, it is important to explore potential mechanisms that may explain how frequent opium use could increase the risk of CRC. One potential mechanism that was considered a priori is constipation, which is a known risk factor for CRC (Sundbøll et al. 2019). It is possible that lead, which is sometimes added to opium as an adulterant, may contribute to constipation (Hayatbakhsh et al. 2017). A study by Nemati et al. demonstrated that blood lead levels were three times higher in opium users than in those who did not use opium (Nemati et al. 2016). While frequent opium use may increase the risk of constipation, this study's findings suggest that constipation may not have a significant impact on the risk of CRC.

It has been reported that some heavy metals used as additives to opium, including lead, arsenic, sulfate, chromium, and cadmium, have been suggested as possible risk factors for CRC (Hayatbakhsh et al. 2017; IARC Monographs Vol 126 group 2020). One plausible mechanism for this increased risk may be due to DNA damage caused by exposure to dichromate ions, which can induce DNA methylation and gene silencing (Bodmer et al. 1987; Karstensen et al. 2019; Wu et al. 2021). A study conducted by Sohrabi et al.

demonstrated that the average concentration of lead in CRC tissues was approximately two times higher than in healthy tissues (Sohrabi et al. 2018). Another study by Etemadi et al. suggested that long-term regular opium use may lead to an increased risk of cancer due to the presence of lead in the blood (Etemadi et al. 2022). Unfortunately, this study did not include information on the intake of heavy metals by the cases and controls, so it was not possible to directly assess the effect of heavy metals on the risk of CRC.

Although there is an association between the frequency of opium use and the risk of CRC, an association between the cumulative count of opium use and the risk of CRC would be expected. However, no such association was observed in this study. The OR did not depend on the age at which opium use began, the length of time since opium use was stopped, or the number of years of opium use. Additional years of opium use appeared to reduce, rather than increase, the risk. This could be explained by the possibility that opium users with constipation problems cannot continue using opium for a long time.

Wu et al. conducted research indicating that the utilization of opium is associated with the development of periodontal disease (Wu et al. 2021). Periodontal diseases may raise systemic inflammation, cause complications with the immune system, modify gut microbiota, and potentially impact the development of CRC (Momen-Heravi et al. 2017). The data analysis revealed that the quantity of decayed teeth used as an indicator of oral hygiene did not demonstrate a significant impact on the risk of developing CRC.

Another possible explanation for the association between regular opium use and an elevated risk of CRC could be linked to polyps, which are a known risk factor for CRC (Bodmer et al. 1987; Karstensen et al. 2019). If opium smoking has a similar effect on the risk of polyps as demonstrated by cigarette smoking (Botteri et al. 2020), it is conceivable that opium use could indirectly raise the risk of developing CRC. Although this study lacked data on polyps, it was recommended that future research be conducted to investigate the potential impact of opium use on the development of polyposis.

This study revealed that there is an additive interaction between opium use and tobacco use, meaning that the combined effect of using both substances is greater than the sum of their individual effects. This finding is consistent with previous studies (Cumberbatch et al. 2018; Sadeghi, Behmard, and Vesselinovitch 1979), as previous studies have also reported a 2-fold increase in the risk of BC for tobacco use alone, while regular users of both opium and tobacco showed a 7-fold increase in the risk of developing BC. There is one study that has suggested a combined effect of opium use and cigarette smoking on the risk of BC (Sadeghi, Behmard, and Vesselinovitch 1979). In that study, the interaction between opium use and cigarette smoking was found to be multiplicative, but the analysis was based on only one case of BC who had used opium exclusively. Additionally, this doctoral thesis found an additive interaction between opium use and tobacco use and the risk of HNSCC.

It is conceivable that some individuals may have begun using opium as a means of alleviating pain associated with cancer symptoms. However, this study utilized a three-year lag period in the analysis, which means that opium use within the three years preceding the interview was not included in the analysis of the data on opium users. This approach helps mitigate any potential bias associated with reverse causality, and therefore, these findings should not be influenced by this bias. It is possible that the risk of reverse causality bias could be low even without the three-year lag assumption, as there was only a small number of individuals (11 cases and 17 controls) who reported beginning opium use less than three years before the interview. Nonetheless, the use of a lag period in this analysis provides an additional level of assurance

that the results are not confounded by reverse causality. The majority of opium users in both the case and control groups had been using opium for over 20 years.

A major source of error in estimating illegal drug use or other sensitive issues is self-report bias. Previous studies indicate that 30 to 70 per cent of individuals who tested positive for illicit drugs in urine screenings may not report their drug use. The degree of self-report bias can vary depending on the study population (Tourangeau and Yan 2007). Therefore, when conducting case-control studies, something that should be taken into consideration is that hospitalized patients may be more cooperative than healthy individuals as a control group. Hospitalized patients may be more likely to report their drug use patterns, which could lead to less self-report bias compared to using healthy individuals as controls (Shuster and Cook 1983; Zhao, Stockwell, and Macdonald 2009). However, the IROPICAN team demonstrated that there was no difference in the sensitivity of self-reporting opium use between hospitalized patients and healthy visitors (Rashidian et al. 2017). Most previous studies that compare self-reported drug use to urine analyses have shown that self-reporting has high specificity, generally above 90%. However, the sensitivity of self-reporting varies widely, ranging from 40% to 75%, depending on the study population and the type of drug used. This means that individuals who use drugs may be less likely to report their use accurately and that urine analyses may be more reliable for detecting drug use in research studies (Harrison et al. 2007). Therefore, it is reasonable to assume that false positives are relatively rare in the self-reporting of drug use, whereas false negatives may be more common.

One of the main challenges in conducting observational studies on the impact of opium use is obtaining accurate, valid and reliable data from both cases and controls since opium use is considered a criminal offence and carries a social stigma. This could lead to misclassification bias. However, previous studies have demonstrated that the self-reporting of opium use was equally sensitive among both cases and controls, which helps alleviate concerns of bias in our study (Hadji et al. 2021; Rashidian et al. 2017).

This study had several notable strengths, including large sample size, histologic confirmation of all cancer cases, and the use of healthy hospital visitor controls. This is in contrast to other hospital-based case-control studies where the controls had other illnesses or conditions, which may confound the analysis. By using healthy hospital visitors as controls, it was possible to minimize the potential for bias due to confounding by underlying health conditions (Rashidian et al. 2017). Furthermore, the quality of data in this study is high because it was collected by trained interviewers using a validated questionnaire (Hadji et al. 2021). This helps ensure that the information gathered is accurate and reliable. Validated questionnaires are rigorously tested to ensure that they accurately capture relevant data, and trained interviewers are able to guide participants through the questionnaire and clarify any ambiguities, further improving the quality of the data collected. Having access to detailed data on the amount of opium used over time allowed us to explore the dose-response relationship between opium use and cancer, as well as the effects of the timing of opium use. There was also comprehensive information on the primary confounder, tobacco, which was taken into account in the statistical models. Additionally, the response rate among both cancer cases and controls was high. However, despite the efforts to control for potential confounding variables, unknown or unmeasured confounders may have affected the results. Additionally, there may be residual confounding from the measured variables that cannot be completely accounted for in the analysis.

7 CONCLUSIONS

The result of this project strongly suggests that opium use is associated with an increased risk of HNSCC, BC, and CRC. The harmful effects of opium on the human body, such as its carcinogenic properties and potential to disrupt DNA integrity, underscore the need for comprehensive public health strategies to address this issue. Efforts should focus on raising awareness about the dangers of opium use, implementing preventive measures, and providing support for individuals struggling with opium addiction. By taking proactive steps to minimize opium consumption and promote healthier lifestyles, it is possible to strive towards reducing the burden of cancer in the communities.

8 FUTURE RESEARCH NEEDS AND OPPORTUNITIES

Further research should clarify the nature and extent of the association between opium use and cancer, which could have important implications for public health and cancer prevention efforts. Future research could focus on isolating the different components of opium and determining which ones are responsible for the observed increase in cancer risk. Researchers should investigate the possible biological mechanisms by which opium use might contribute to cancer development, such as oxidative stress, inflammation, or DNA damage.

Future studies could explore the relationship between opium use and the risk of different types of cancer including cancers of the esophagus, stomach, pharynx, and pancreas should be investigated further in well-conducted studies. Researchers should examine whether opium use interacts with other known risk factors of cancer, such as smoking or exposure to environmental agents. Longitudinal studies would be needed to follow opium users over time and track their cancer risk, as well as identify any factors that may modify the association between opium use and cancer risk. The research on the carcinogenic mechanisms of opium use remains unclear, leaving a vast untapped potential for further exploration and investigation in this field.

In addition, future research on the association between opioid use and cancer is critical due to the increasing prevalence of opioid use and the need to comprehend potential long-term health implications. Understanding whether there is any association, identifying risk factors, and finding the mechanisms that can significantly impact public health policies, clinical practices, and patient outcomes is also key.

The findings of future research should be used as the basis for developing prevention strategies to reduce the risk of cancer related to opium use. These strategies could include public health campaigns to raise awareness of the potential risks of opium use, as well as interventions to help people quit or reduce their opium use.

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10 REFERENCES

Abdolahinia, Z., Pakmanesh, H. , Mirzaee, M., Bazrafshan, A., Shafiei Bafti, M., and Shahesmaeili, A. 2021. "Opium and Cigarette Smoking Are Independently Associated with Bladder Cancer: The Findings of a Matched Case-Control Study." *Asian Pacific Journal of Cancer Prevention: APJCP* 22 (10): 3385.

Adcock, J. J. 1991. "Peripheral Opioid Receptors and the Cough Reflex." *Respiratory Medicine* 85: 43–46.

Afshari, M., Janbabaei, G., Bahrami, M.A., and Moosazadeh, M. 2017. "Opium and Bladder Cancer: A Systematic Review and Meta-Analysis of the Odds Ratios for Opium Use and the Risk of Bladder Cancer." *PloS One* 12 (6): e0178527.

Akbari, M., Naghibzadeh-Tahami, A., Khanjani, N., Baneshi, M.R., Kamali, E., Hesampour, M., et al. 2015. "Opium as a Risk Factor for Bladder Cancer: A Population-Based Case-Control Study in Iran." *Archives of Iranian Medicine* 18 (9): 567–71.

Al Hussein Al Awamlh, B., Shoag, J.E., Ravikumar, V., Posada, L., Taylor, B.L., van der Mijn, J.C., et al. 2019. "Association of Smoking and Death from Genitourinary Malignancies: Analysis of the National Longitudinal Mortality Study." *The Journal of Urology* 202 (6): 1248–54.

Aliramaji, A., Kaseean, A., Yousefnia Pasha,Y.R, Shafi, H., Kamali, S., Safari, M., et al. 2015. "Age Distribution Types of Bladder Cancers and Their Relationship with Opium Consumption and Smoking." *Caspian Journal of Internal Medicine* 6 (2): 82.

Alizadeh, H., Naghibzadeh-Tahami, A., Khanjani, N., Yazdi-Feyzabadi, V., Eslami, H., Borhaninejad, V., et al. 2020. "Opium Use and Head and Neck Cancers: A Matched Case-Control Study in Iran." *Asian Pacific Journal of Cancer Prevention: APJCP* 21 (3): 783.

Viale PH. The American Cancer Society's Facts & Figures: 2020 Edition. *J Adv Pract Oncol*. 2020 Mar;11(2):135-136. doi: 10.6004/jadpro.2020.11.2.1. Epub 2020 Mar 1. PMID: 33532112; PMCID: PMC7848816.

Amin-Esmaeili, M., Rahimi-Movaghar, A., Sharifi, V., Hajebi, A., Radgoodarzi, R., Mojtabai, R., et al. 2016. "Epidemiology of Illicit Drug Use Disorders in Iran: Prevalence, Correlates, Comorbidity and Service Utilization Results from the Iranian Mental Health Survey." *Addiction* 111 (10): 1836–47.

Andersson, T., Alfredsson, L., Källberg, H., Zdravkovic, S., and Ahlbom, A. 2005. "Calculating Measures of Biological Interaction." *European Journal of Epidemiology* 20: 575–79.

Asgari, A., Kaviani, A., Gachkar, L., and Hosseini-Nassab, S. R. 2004. "Is Bladder Cancer More Common among Opium Addicts?" *Urology Journal* 1 (4): 253–55.

Auguste, A., Deloumeaux, J., Joachim, C., Gaete, S., Michineau, L., Herrmann-Storck, C., et al. 2020. "Joint Effect of Tobacco, Alcohol, and Oral HPV Infection on Head and Neck Cancer Risk in the French West Indies." *Cancer Medicine* 9 (18): 6854–63.

Aupérin, A. 2020. "Epidemiology of Head and Neck Cancers: An Update." *Current Opinion in Oncology* 32 (3): 178–86.

Aveta, A., Cacciapuoti, C., Barone, B., Di Zazzo, E., Del Giudice, F., Maggi, M., et al. 2022. "The Impact of Meat Intake on Bladder Cancer Incidence: Is It Really a Relevant Risk?" *Cancers* 14 (19): 4775.

Baan, R., Grosse, Y., Straif, K., Secretan, B., El Ghissassi, F., Bouvard, V., Benbrahim-Tallaa, L., et al. 2009. "A Review of Human Carcinogens. F. Chemical Agents and Related Occupations: IARC Monographs on the Evaluation of Carcinogenic Risks to Humans." Lancet Oncol 10 (12): 1143–44. *https://doi.org/10.1016/s1470-2045(09)70358-4*.

Bakhshaee, M., Raziee, H.R., Afshari, R., Amali, A., Roopoosh, M., and Lotfizadeh, A. 2017. "Opium Addiction and Risk of Laryngeal and Esophageal Carcinoma." *Iranian Journal of Otorhinolaryngology* 29 (90): 19.

Beattie, A. D., Briggs, J. D., Canavan, J. S. F., Doyle, D., Mullin, P. J., and Watson, A. A. 1975. "Acute Lead Poisoning: Five Cases Resulting from Self-Injection of Lead and Opium." *QJM: An International Journal of Medicine* 44 (2): 275–84. https://doi.org/10.1093/oxfordjournals.qjmed.a067425.

Bertazzi, P. A., Consonni, D., Bachetti, S., Rubagotti, M., Baccarelli, A., Zocchetti, C., et al. 2001. "Health Effects of Dioxin Exposure: A 20-Year Mortality Study." *American Journal of Epidemiology* 153 (11): 1031–44.

Bidary, M. Z., Sahranavard, M., Akhavan Rezayat, A., Omranzadeh, A., Hoseiny, S.H., Kabirian, A., et al. 2021. "Opium as a Carcinogen: A Systematic Review and Meta-Analysis." *EClinicalMedicine* 33: 100768.

Bodmer, W. F., Bailey, C.J., Bodmer, J., Bussey, H.J.R., Ellis, A., Gorman, P., et al. 1987. "Localization of the Gene for Familial Adenomatous Polyposis on Chromosome 5." *Nature* 328 (6131): 614–16.

Botteri, E., Borroni, E., Sloan, E.K., Bagnardi,V., Bosetti, C., Peveri, G., et al. 2020. "Smoking and Colorectal Cancer Risk, Overall and by Molecular Subtypes: A Meta-Analysis." *Official Journal of the American College of Gastroenterology | ACG* 115 (12): 1940–49.

Calvert, G. M., Ward, E., Schnorr, T. M., and Fine, L. J. 1998. "Cancer Risks among Workers Exposed to Metalworking Fluids: A Systematic Review." *American Journal of Industrial Medicine* 33 (3): 282–92.

Canda, M. Ş., Eroğlu, O. N., and Hapa, O. 2021. "International Classification of Diseases for Oncology (ICD-O) Coding System, Language for Oncology Implications and Update at Orthopaedic Oncology." *Turkish Journal of Oncology/Türk Onkoloji Dergisi* 36 (2).

Chang, J. S., Lo, H., Wong, T., Huang, C., Lee,W., Tsai, S., et al. 2013. "Investigating the Association between Oral Hygiene and Head and Neck Cancer." *Oral Oncology* 49 (10): 1010–17.

Chaturvedi, A. K., Anderson, W.F., Lortet-Tieulent, J., Curado, M. P., Ferlay, J., Franceschi, S., et al. 2013. "Worldwide Trends in Incidence Rates for Oral Cavity and Oropharyngeal Cancers." *Journal of Clinical Oncology* 31 (36): 4550.

Chuang, S., Jenab, M., Heck, J.E. , Bosetti, C., Talamini, R., Matsuo, K., et al. 2012. "Diet and the Risk of Head and Neck Cancer: A Pooled Analysis in the INHANCE Consortium." *Cancer Causes & Control* 23: 69–88.

Conway, D. I., Brenner, D.R., McMahon, A. D., Macpherson, L., Agudo, A., Ahrens, W., et al. 2015. "Estimating and Explaining the Effect of Education and Income on Head and Neck Cancer Risk: INHANCE Consortium Pooled Analysis of 31 Case-Control Studies from 27 Countries." *International Journal of Cancer* 136 (5): 1125–39.

Cumberbatch, M. G. K., Jubber, I., Black, P. C., Esperto, F., Figueroa, J. D., Kamat, A. M., et al. 2018. "Epidemiology of Bladder Cancer: A Systematic Review and Contemporary Update of Risk Factors in 2018." *European Urology* 74 (6): 784–95.

De Rosa, M., Pace, U., Rega, D., Costabile, V., Duraturo, F., Izzo, P., et al. 2015. "Genetics, Diagnosis and Management of Colorectal Cancer." *Oncology Reports* 34 (3): 1087–96.

Deb, A. A., Okechukwu, C. E., Emara, S. , and Sami, A. A. 2019. "Occupational Exposure as Risk Factor for Kidney and Bladder Cancer: A Systematic Review and Meta-Analysis." *Urol. Nephrol. Open Access J* 7: 143–51.

Dekker, E. , Tanis, P.J., Vleugels, J.L.A., Kasi, P.M., and Wallace, M.B. 2019. "Colorectal Cancer" 394(10207) (October): 1467–80. https://doi.org/10.1016/S0140-6736(19)32319-0.

Dement, J., Pompeii, L., Lipkus, I. M., and Samsa, G.P. 2003. "Cancer Incidence among Union Carpenters in New Jersey." *Journal of Occupational and Environmental Medicine*, 1059–67.

Di Credico, G., Edefonti, V., Polesel, J., Pauli, F., Torelli, N., Serraino, D., et al. 2019. "Joint Effects of Intensity and Duration of Cigarette Smoking on the Risk of Head and Neck Cancer: A Bivariate Spline Model Approach." *Oral Oncology* 94: 47–57.

Dianatinasab, M., Wesselius,A., Loeij, T., Salehi-Abargouei, A., YW Yu, E., Fararouei, M., et al. 2021. "The Association between Meat and Fish Consumption and Bladder Cancer Risk: A Pooled Analysis of 11 Cohort Studies." *European Journal of Epidemiology* 36 (8): 781–92.

Divaris, K., Olshan, A.F., Smith, J., Bell, M.E., Weissler, M.C., Funkhouser, W.K., et al. 2010. "Oral Health and Risk for Head and Neck Squamous Cell Carcinoma: The Carolina Head and Neck Cancer Study." *Cancer Causes & Control* 21: 567–75.

U.S. Department of Justice, Drug Enforcement Administration (DEA). 1992. "Opium Poppy Cultivation and Heroin Processing in Southeast Asia." *Washington DC: Office of Intelligence.*, 20537 (202): 307-8100

Dumas, S., Parent, M., Siemiatycki, J., and Brisson, J. 2000. "Rectal Cancer and Occupational Risk Factors: A Hypothesis-Generating, Exposure-Based Case-Control Study." *International Journal of Cancer* 87 (6): 874–79.

Edefonti, V., Garavello, W., La Vecchia, C., Parpinel, M., Franceschi, S., Dal Maso, L., et al. 2010. "Nutrient-Based Dietary Patterns and Laryngeal Cancer: Evidence from an Exploratory Factor Analysis." *Cancer Epidemiology, Biomarkers & Prevention* 19 (1): 18-27 (a).

Edefonti, V., La Vecchia, C., Randi, G., Ferraroni, M., Garavello, Franceschi, W.S., et al. 2010. "Nutrient-Based Dietary Patterns and the Risk of Oral and Pharyngeal Cancer." *Oral Oncology* 46 (5): 343-348 (b).

Elangovan, A., Skeans, Landsman, M., Ali, S.M.J., Elangovan, A. G., Kaelber, D.C., et al. 2021. "Colorectal Cancer, Age, and Obesity-Related Comorbidities: A Large Database Study." *Digestive Diseases and Sciences* 66: 3156–63.

Etemadi, A., Hariri, S., Hassanian-Moghaddam, H., Poustchi, H., Roshandel, G., Shayanrad, A., et al. 2022. "Lead Poisoning among Asymptomatic Individuals with a Long-Term History of Opiate Use in Golestan Cohort Study." *International Journal of Drug Policy* 104 (2022): 103695. https://doi.org//doi.org/10.1016/j.drugpo.2022.103695.

Etminan, M., Collins, G.S., and Mansournia, M.A. 2020. "Using Causal Diagrams to Improve the Design and Interpretation of Medical Research." *Chest* 158 (1): S21–28. https://doi.org/10.1016/j.chest.2020.03.011.

Fahmy, M.S., Sadeghi, A., and Behmard, S. 1983. "Epidemiologic Study of Oral Cancer in Fars Province, Iran." *Community Dentistry and Oral Epidemiology* 11 (1): 50–58.

Farvid, M.S., Sidahmed, E., Spence, N.D., Mante Angua, K., Rosner, B.A., and Barnett, J.B. 2021. "Consumption of Red Meat and Processed Meat and Cancer Incidence: A Systematic Review and Meta-Analysis of Prospective Studies." *European Journal of Epidemiology* 36: 937–51.

Feng, Y., Dai, W., Li, Y., Mo, S., Li, Q., and Cai, S. 2018. "The Effect of Marital Status by Age on Patients with Colorectal Cancer over the Past Decades: A SEER-Based Analysis." *International Journal of Colorectal Disease* 33: 1001–10.

Ferlay, J., Ervik, M., Lam, F., Colombet, M., Mery, L., Piñeros, M., et al. 2023. "Global Cancer Observatory: Cancer Today. International Agency for Research on Cancer." Lyon, France. 2023.

Ferrari, P., Jenab, M., Norat, T., Moskal, A., Slimani, N., Olsen, A., et al. 2007. "Lifetime and Baseline Alcohol Intake and Risk of Colon and Rectal Cancers in the European Prospective Investigation into Cancer and Nutrition (EPIC)." *International Journal of Cancer* 121 (9): 2065–72.

Filho, A., Turner, M.C., Warnakulasuriya, S., Richardson, D.B., Hosseini, B., Kamangar, F., et al. 2023. "The Carcinogenicity of Opium Consumption: A Systematic Review and Meta-Analysis." *European Journal of Epidemiology*, 1–17.

Freedman, Neal D., Silverman, D. T., Hollenbeck, A. T., Schatzkin, A., and Abnet, C. 2011. "Association between Smoking and Risk of Bladder Cancer among Men and Women." *Jama* 306 (7): 737–45.

Friesen, M., I. K. O'neill, C. Malaveille, L. Garren, A. Hautefeuille, J. R. P. Cabral, et al. 1985. "Characterization and Identification of 6 Mutagens in Opium Pyrolysates Implicated in Oesophagel Cancer in Iran." *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis* 150 (1–2): 177–91.

Fritz, A., Percy, C., Jack, A., Shanmugaratnam, K., Sobin, L., Parkin, D., et al. 2019. "International Classification of Diseases for Oncology (ICD-O), 2nd Rev." *World Health Organization: Geneva, Switzerland*.

Garabrant, D.H., Peters, J.M., Mack, T.M., and Bernstein, L. 1984. "Job Activity and Colon Cancer Risk." *American Journal of Epidemiology* 119 (6): 1005–14.

Ghadimi, T., Gheitasi, B., Nili, S., Karimi, M., and Ghaderi, E. 2015. "Occupation, Smoking, Opium, and Bladder Cancer: A Case–Control Study." *South Asian Journal of Cancer* 4 (03): 111–14.

Ghadirian, P., Stein, G.F., Gorodetzky, C. , Roberfroid, M.B., Mahon, G.A.T., Bartsch, H., et al. 1985. "Oesophageal Cancer Studies in the Caspian Littoral of Iran: Some Residual Results, Including Opium Use as a Risk Factor." *International Journal of Cancer* 35 (5): 593–97.

Grandhi, R.K., Lee, S., and Abd-Elsayed, A. 2017. "Does Opioid Use Cause Angiogenesis and Metastasis?" *Pain Medicine* 18 (1): 140–51.

Grimes, D.A., and Schulz, K.F. 2005. "Compared to What? Finding Controls for Case-Control Studies." *The Lancet* 365 (9468): 1429–33.

Hadji, M., Rashidian, H., Marzban, M., Gholipour, M., Naghibzadeh-Tahami, A., Mohebbi, E., et al. 2021. "The Iranian Study of Opium and Cancer (IROPICAN): Rationale, Design, and Initial Findings." *Arch Iran Med* 24 (3): 167–76. https://doi.org/10.34172/aim.2021.27.

Hadji, M., Rashidian, H., Marzban, M., Naghibzadeh-Tahami, A., Gholipour, M., Mohebbi, E., et al. 2022. "Opium Use and Risk of Bladder Cancer: A Multicenter Case-Referent Study in Iran." *International Journal of Epidemiology* 51 (3): 830–38.

Hampel, H., Frankel, W. L., Martin, E:, Arnold, M., Khanduja, K., Kuebler, P., et al. 2005. "Screening for the Lynch Syndrome (Hereditary Nonpolyposis Colorectal Cancer)." *New England Journal of Medicine* 352 (18): 1851–60. https://doi.org/10.1056/NEJMoa043146.

Harrison, L. D., Martin, S., Enev, T., and Harrington, D.. 2007. "Comparing Drug Testing and Self-Report of Drug Use among Youths and Young Adults in the General Population." *Rockville, MD: Substance Abuse and Mental Health Services Administration, Office of Applied Studies*.

Hasanpour-Heidari, S., Fazel, A., Semnani, S., Khandoozi, S.R., Amiriani, T., Sedaghat, S.M., et al. 2019. "Temporal and Geographical Variations in Colorectal Cancer Incidence in Northern Iran 2004–2013." *Cancer Epidemiology* 59: 143–47.

Hashibe, M., Brennan, P., Benhamou, S., Castellsague, X., Chen, C., Paula Curado, M., et al. 2007. "Alcohol Drinking in Never Users of Tobacco, Cigarette Smoking in Never Drinkers, and the Risk of Head and Neck Cancer: Pooled Analysis in the International Head and Neck Cancer Epidemiology Consortium." *Journal of the National Cancer Institute* 99 (10): 777–89.

Hashibe, M., Hunt, J., Wei, M., Buys, S., Gren, L., and Lee, Y. A. 2013. "Tobacco, Alcohol, Body Mass Index, Physical Activity, and the Risk of Head and Neck Cancer in the Prostate, Lung, Colorectal, and Ovarian (PLCO) Cohort." *Head & Neck* 35 (7): 914–22.

Hashim, D., Sartori, S. , Brennan, P., Curado, M. P., Wünsch-Filho, V., Divaris, K., et al. 2016. "The Role of Oral Hygiene in Head and Neck Cancer: Results from International Head and Neck Cancer Epidemiology (INHANCE) Consortium." *Annals of Oncology* 27 (8): 1619–25.

Hashim, D., Sartori, S., Vecchia,C. L., Serraino, D., Dal Maso, L., Negri, E., et al. 2017. "Hormone Factors Play a Favorable Role in Female Head and Neck Cancer Risk." *Cancer Medicine* 6 (8): 1998–2007.

Hayatbakhsh, M.M., Oghabian, Z., Conlon, E., Nakhaee, S., Amirabadizadeh, A.R., Zahedi, M. J., et al. 2017. "Lead Poisoning among Opium Users in Iran: An Emerging Health Hazard." *Substance Abuse Treatment, Prevention, and Policy* 12 (1): 1–8. https://doi.org/10.1186/s13011-017-0127-0.

Hewer, T., Rose, E., Ghadirian, P., Castegnaro, M., Malaveille, C., Bartsch, H., et al. 1978. "Ingested Mutagens from Opium and Tobacco Pyrolysis Products and Cancer of the Oesophagus." *The Lancet* 312 (8088): 494–96.

Hosseini, S.Y., Safarinejad, M.R., Amini, E., and Hooshyar, H. 2010. "Opium Consumption and Risk of Bladder Cancer: A Case-Control Analysis." In *Urologic Oncology: Seminars and Original Investigations*, 28:610–16. Elsevier.

"Https://Nordcan.Iarc.Fr/En." 2023. February 20, 2023.

IARC. 2006. "Monographs on the Evaluation of Carcinogenic Risks to Humans." *Http://Monographs. Iarc. Fr/ENG/Classification/Index. Php*.

IARC. 2023. "Global Cancer Observatory". https://gco.iarc.fr, (February25).

IARC Monographs Vol 126 group. 2020. "Carcinogenicity of Opium Consumption." *The Lancet. Oncology* 21 (11): 1407–8. https://doi.org/10.1016/S1470-2045(20)30611-2.

Jakszyn, P., González,C. A., Luján-Barroso, L., Ros, M., Bueno-de-Mesquita, H.B., Roswall, N., et al. 2011. "Red Meat, Dietary Nitrosamines, and Heme Iron and Risk of Bladder Cancer in the European Prospective Investigation into Cancer and Nutrition (EPIC)." *Cancer Epidemiology, Biomarkers & Prevention* 20 (3): 555–59.

Jess, T., Riis, L., Vind, I., Vanessa Winther, K., Borg, S., Binder, V., et al. 2007. "Changes in Clinical Characteristics, Course, and Prognosis of Inflammatory Bowel Disease during the Last 5 Decades: A Population-Based Study from Copenhagen, Denmark." *Inflammatory Bowel Diseases* 13 (4): 481–89.

Johnson, D.E., Burtness, B., Leemans, C.R., Wai Yan Lui, V., Bauman, J E., and Grandis, J. R. 2020. "Head and Neck Squamous Cell Carcinoma." *Nature Reviews Disease Primers* 6 (1): 1–22.

Kalan-Farmanfarma, K., Mahdavifar, N., and Salehiniya, H. 2020. "Bladder Cancer in Iran: An Epidemiological Review." *Research and Reports in Urology*, 91–103.

Kalant, H. 1997. "Opium Revisited: A Brief Review of Its Nature, Composition, Non-Medical Use and Relative Risks 1." *Addiction* 92 (3): 267–77.

Kamangar, F., Shakeri, R., Malekzadeh, R., and Islami, F. 2014. "Opium Use: An Emerging Risk Factor for Cancer?" *The Lancet Oncology* 15 (2): e69–77.

Karstensen, J.G., Burisch, J., Pommergaard, H.C, Aalling, L., Højen, H., Jespersen, N., et al. 2019. "Colorectal Cancer in Individuals with Familial Adenomatous Polyposis, Based on Analysis of the Danish Polyposis Registry." *Clinical Gastroenterology and Hepatology* 17 (11): 2294–2300. https://doi.org/10.1016/j.cgh.2019.02.008.

Kauppinen, T., Toikkanen, J., and Pukkala, E. 1998. "From Cross-Tabulations to Multipurpose Exposure Information Systems: A New Job-Exposure Matrix." *American Journal of Industrial Medicine* 33 (4): 409–17.

Kauppinen, T., Uuksulainen, S., Saalo, A., Mäkinen, I., and Pukkala, E. 2014. "Use of the Finnish Information System on Occupational Exposure (FINJEM) in Epidemiologic, Surveillance, and

Other Applications." *Annals of Occupational Hygiene* 58 (3): 380–96. https://doi.org/10.1093/annhyg/met074.

Kerber, R.A., Slattery, M.L., Potter, J.D., Caan, B.J., and Edwards, S.L. 1998. "Risk of Colon Cancer Associated with a Family History of Cancer or Colorectal Polyps: The Diet, Activity, and Reproduction in Colon Cancer Study." *International Journal of Cancer* 78 (2): 157–60.

Keshtkar, A.A., Semnani, S., Pourshams, A., Khademi, H., Roshandel, G., Boffetta, P., et al. 2010. "Pictogram Use Was Validated for Estimating Individual's Body Mass Index." *Journal of Clinical Epidemiology* 63 (6): 655–59. https://doi.org/10.1016/j.jclinepi.2009.08.014.

Ketabchi, A., Gharaei, M., Ahmadnezhad, M., and Mirshekari, T. 2005. "Evaluation of Bladder Cancer in Opium Addicted Patients in the Kerman Province, Iran, from 1999 to 2003." *Journal of Research in Medical Sciences* 10 (6): 355–57.

Kewenter, J.,H. Ahlman, and Hulten, L. 1978. "Cancer Risk in Extensive Ulcerative Colitis." *Annals of Surgery* 188 (6): 824.

Khademi, H., Malekzadeh, R., Pourshams, A., Jafari, E., Salahi, R., Semnani, S., et al. 2012. "Opium Use and Mortality in Golestan Cohort Study: Prospective Cohort Study of 50 000 Adults in Iran." *British Medical Journal* 344 (e2502): 1–12.

Kheirandish, P., Seyedalinaghi, S.A., Hosseini, M., Jahani, M.R., Shirzad, H., Foroughi, M., et al. 2010. "Prevalence and Correlates of HIV Infection among Male Injection Drug Users in Detention in Tehran, Iran." *JAIDS Journal of Acquired Immune Deficiency Syndromes* 53 (2): 273–75.

Khetan, P., Boffetta, P., Luce, D., Stucker, I., Curado, M.P., Menezes, A., et al. 2019. "Occupations and the Risk of Head and Neck Cancer: A Pooled Analysis of the International Head and Neck Cancer Epidemiology (INHANCE) Consortium." *Journal of Occupational and Environmental Medicine* 61 (5): 397.

Khoo, R. 1981. "Radiotherapy of Carcinoma of the Larynx." *Annals of the Academy of Medicine, Singapore* 10 (3): 307–10.

Khosravizadegan, Z., Naghibzadeh-Tahami, A., Akbari, M., Khodadost, M., Honarvar, B., Khodadost, B., et al. 2017. "Opium Use and Risk of Lower Gastrointestinal Cancers: Population-Based Case-Control Study in South of Iran." *International Journal of Cancer Management* 10 (6). https://doi.org/10.5812/ijcm.8227.

Kim, S., Young Paik, H., Yoon, H., Eun Lee, J., Kim, N. and Sung, M. 2015. "Sex-and Gender-Specific Disparities in Colorectal Cancer Risk." *World Journal of Gastroenterology: WJG* 21 (17): 5167.

Koo, T.K., and Li, M.Y. 2016. "A Guideline of Selecting and Reporting Intraclass Correlation Coefficients for Reliability Research." *Journal of Chiropractic Medicine* 15 (2): 155–63.

Kossenas, K., and Constantinou, C. 2021. "Epidemiology, Molecular Mechanisms, and Clinical Trials: An Update on Research on the Association between Red Meat Consumption and Colorectal Cancer." *Current Nutrition Reports*, 1–33.

Labanca, F., Ovesna, J. and Milella, L. 2018. "Papaver Somniferum L. Taxonomy, Uses and New Insight in Poppy Alkaloid Pathways." *Phytochemistry Reviews* 17: 853–71.

Lagiou, P., Talamini, R., Samoli, E., Lagiou, A., Ahrens, W., Pohlabeln, H., et al. 2009. "Diet and Upper-Aerodigestive Tract Cancer in Europe: The ARCAGE Study." *International Journal of Cancer* 124 (11): 2671–76.

Larsson, S. C., Carter, P., Kar, S., Vithayathil, M., Mason, A.M., Michaëlsson, K., et al. 2020. "Smoking, Alcohol Consumption, and Cancer: A Mendelian Randomisation Study in UK Biobank and International Genetic Consortia Participants." *PLoS Medicine* 17 (7): e1003178.

Lenis, A.T., Lec, P.M., and Chamie, K. 2020. "Bladder Cancer: A Review." *Jama* 324 (19): 1980–91.

Lewallen, S., and Courtright, P. 1998. "Epidemiology in Practice: Case-Control Studies." *Community Eye Health* 11 (28): 57.

Li, H., Chen, X., Hoffmeister, M., and Brenner, H. 2023. "Associations of Smoking with Early-and Late-Onset Colorectal Cancer." *JNCI Cancer Spectrum* 7 (1): pkad004.

Lin, C., Lee, W., Ou, C., Hsiao, J., Huang, C., Huang, J., et al. 2017. "Regular Recreational Physical Activity and Risk of Head and Neck Cancer." *BMC Cancer* 17 (1): 1–10.

Linn, J. F., Sesterhenn, I., Mostofi, F.K., and Schoenberg, M. 1998. "The Molecular Characteristics of Bladder Cancer in Young Patients." *The Journal of Urology* 159 (5): 1493–96.

Liu, P., Wu, K., Ng, K., Zauber, A.G., Nguyen, L H., Song, M., et al. 2019. "Association of Obesity with Risk of Early-Onset Colorectal Cancer among Women." *JAMA Oncology* 5 (1): 37–44.

Lotfi, M., Farzaneh, F., Mehrparvar, A.H., Fallahzadeh, M.H, and Sadeghian, M.R. 2016. "The Effect of Smoking and Opium on Bladder Cancer in Yazd Province: A Case-Control Study." *Journal of Community Health Research* 5 (2): 98–109.

Lutgens, M., Oijen, M., Heijden, G., Vleggaar, F., Siersema, P., and Oldenburg, B. 2013. "Declining Risk of Colorectal Cancer in Inflammatory Bowel Disease: An Updated Meta-Analysis of Population-Based Cohort Studies." *Inflammatory Bowel Diseases* 19 (4): 789–99.

MacLennan, R., Day, J., Law, C., Ng, Y., and Shanmugaratnam, K. 1977. "Risk Factors for Lung Cancer in Singapore Chinese, a Population with High Female Incidence Rates." *International Journal of Cancer* 20 (6): 854–60.

Malekzadeh, M.M., Khademi, H., Pourshams, A., Etemadi, A., Poustchi, H., Bagheri, M., et al. 2013. "Opium Use and Risk of Mortality from Digestive Diseases–a Prospective Cohort Study." *The American Journal of Gastroenterology* 108 (11): 1757.

Mansouri, M., Naghshi, S., Parsaeian, M., Sepanlou, S., Poustchi, H., Momayez Sanat, Z., et al. 2022. "Opium Use and Cancer Risk: A Comprehensive Systematic Review and Meta-Analysis of Observational Studies." *International Journal of Clinical Practice* 2022.

Marley, A.R., and Nan, H. 2016. "Epidemiology of Colorectal Cancer." *International Journal of Molecular Epidemiology and Genetics* 7 (3): 105.

Masjedi, M.R., Adimi Naghan, P., Taslimi, S., Yousefifard, M., Ebrahimi, S.M., Khosravi, A., et al. 2013. "Opium Could Be Considered an Independent Risk Factor for Lung Cancer: A Case-Control Study." *Respiration* 85 (2): 112–18.

Masoudi, M., Zali, M.R., Ehsani, M.J., Mohammadreza, A., Aiassofi, K., Aghazadeh, R., et al. 2006. "Abdominal Pain Due to Lead-Contaminated Opium: A New Source of Inorganic Lead Poisoning in Iran."

Mastrangelo, G., Fedeli, U., Fadda, E., Milan, G., and Lange, J. . 2002. "Epidemiologic Evidence of Cancer Risk in Textile Industry Workers: A Review and Update." *Toxicology and Industrial Health* 18 (4): 171–81.

Matson, S.C. 2011. "Substance Abuse and Dependence." *Adolescent Medicine Today: A Guide To Caring For The Adolescent Patient*, 291.

Mattiuzzi, C., and Lippi, G. 2021. "Epidemiologic Burden of Red and Processed Meat Intake on Colorectal Cancer Mortality." *Nutrition and Cancer* 73 (4): 562–67.

McNabb, S., Harrison, T., Albanes, D., Berndt, S., Brenner, H., Caan, B., et al. 2020. "Meta-Analysis of 16 Studies of the Association of Alcohol with Colorectal Cancer." *International Journal of Cancer* 146 (3): 861–73.

Mehanna, H., Paleri, V., West, C.M.L., and Nutting, C. 2010. "Head and Neck Cancer—Part 1: Epidemiology, Presentation, and Prevention." *British Medical Journal* 341 (c4684): 663–66.

Mehta, S., Arroyave, W., Lunn, R., Mark Park, Y., Boyd, W., and Sandler, D. 2020. "A Prospective Analysis of Red and Processed Meat Consumption and Risk of Colorectal Cancer in Women." *Cancer Epidemiology, Biomarkers & Prevention* 29 (1): 141–50.

Moeller, K.E., Lee, K.C., and Kissack, J.C. 2008. "Urine Drug Screening: Practical Guide for Clinicians." In *Mayo Clinic Proceedings*, 83:66–76.

Mohebbi, E., Hadji, M., Rashidian, H., Rezaianzadeh, A., Marzban, M., Haghdoost, A.A, et al. 2021. "Opium Use and the Risk of Head and Neck Squamous Cell Carcinoma." *International Journal of Cancer* 148 (5): 1066–76.

Mohebbi, E., Rashidian, H., Naghibzadeh Tahami, A., Haghdoost, A.A., Rahimi-Movaghar, A. , M. S. Seyyedsalehi, et al. 2021. "Opium Use Reporting Error in Case-Control Studies: Neighborhood Controls versus Hospital Visitor Controls." *Medical Journal of the Islamic Republic of Iran* 35 (2021): 60–67. https://doi.org/10.47176/mjiri.35.60.

Momen-Heravi, F., Babic, A., Tworoger, S.S., Zhang, L., Wu, K., Smith-Warner, S. A., et al. 2017. "Periodontal Disease, Tooth Loss and Colorectal Cancer Risk: Results from the Nurses' Health Study." *International Journal of Cancer* 140 (3): 646–52. https://doi.org/10.1002/ijc.30486.

Moossavi, S., Mohamadnejad, M., Pourshams, A., Poustchi, H., Islami, F., Sharafkhah, M., et al. 2018. "Opium Use and Risk of Pancreatic Cancer: A Prospective Cohort StudyOpium Use and Risk of Pancreatic Cancer." *Cancer Epidemiology, Biomarkers & Prevention* 27 (3): 268–73.

Mori, K., Mostafaei, H., Abufaraj, M., Yang, L., Egawa, S., and Shariat, S.F. 2020. "Smoking and Bladder Cancer: Review of the Recent Literature." *Current Opinion in Urology* 30 (5): 720–25.

Mousavi, M., Ahamadi, R., Damghani, M.A, Haghdoust, A.A, and Khamesipour, A. 2003. "Opium and Risk of Laryngeal Cancer." *The Laryngoscope* 113 (11): 1939–43.

Murphy, G., Devesa, S.S., Cross, A.J., Inskip, P.D., McGlynn, K.A., and Cook, M.B. 2011. "Sex Disparities in Colorectal Cancer Incidence by Anatomic Subsite, Race and Age." *International Journal of Cancer* 128 (7): 1668–75.

Naghibzadeh-Tahami, A., Khanjani, N., Yazdi- Feyzabadi, V., Varzandeh, M., and Haghdoost, A.A. 2014. "Opium as a Risk Factor for Upper Gastrointestinal Cancers: A Population-Based Case-Control Study in Iran." *Archives of Iraninan Medicine,*17: 1-6

Naghibzadeh-Tahami, A., Yazdi Feyzabadi, V., Khanjani, N., Ashrafi-Asgarabad, A., Alizaeh, H., Borhaninejad, V.R, et al. 2016. "Can Opium Use Contribute to a Higher Risk of Colorectal Cancers? A Matched Case-Control Study in Iran." *Iranian Journal of Public Health* 45 (10): 1322–31.

Nalini, M., Shakeri, R., Poustchi, H., Pourshams, A., Etemadi, A., Islami, F., et al. 2021. "Long-Term Opiate Use and Risk of Cardiovascular Mortality: Results from the Golestan Cohort Study." *European Journal of Preventive Cardiology* 28 (1): 98–106.

Nasrollahzadeh, D., Kamangar, F., Aghcheli, K., Sotoudeh, M. , Islami, F., Abnet, C.C., et al. 2008. "Opium, Tobacco, and Alcohol Use in Relation to Oesophageal Squamous Cell Carcinoma in a High-Risk Area of Iran." *British Journal of Cancer* 98 (11): 1857–63.

U.S, National Institute of Health, National Cancer Institute." (27. Feb. 2023). *Https://Training.Seer.Cancer.Gov (February 2023). 2023*.

Nemati, A., Jafari, Sh., Afshari, M., Dahmardeh, S., and Tabrizian, K. 2016. "Comparing Blood Lead Level among Oral/Inhaled Opium Addicts with a Non-Addict Control Group in the Southeast of Iran." *Addiction and Health* 8 (4): 235–41.

Nicolotti, N., Chuang, S., Cadoni, G., Arzani, D., Petrelli, L., Bosetti, C., et al. 2011. "Recreational Physical Activity and Risk of Head and Neck Cancer: A Pooled Analysis within the International Head and Neck Cancer Epidemiology (INHANCE) Consortium." *European Journal of Epidemiology* 26: 619–28.

Nourbakhsh, A., and MG Mohseni ZN Hatmi. 2006. "Opium Use in Transitional Cell Carcinoma of the Urinary Bladder." *Acta Medica Iranica*, 263–68.

Oddone, E., Modonesi, C., and Gatta, G. 2014. "Occupational Exposures and Colorectal Cancers: A Quantitative Overview of Epidemiological Evidence." *World Journal of Gastroenterology: WJG* 20 (35): 12431.

Osch, F., Vlaanderen, J., Jochems, S., Bosetti, C., Polesel, J., Porru, S., et al. 2019. "Modeling the Complex Exposure History of Smoking in Predicting Bladder Cancer: A Pooled Analysis of 15 Case–Control Studies." *Epidemiology (Cambridge, Mass.)* 30 (3): 458.

Papadimitriou, N., Dimou, N., Tsilidis, K., Banbury, B., Martin, R., Lewis, S., et al. 2020. "Physical Activity and Risks of Breast and Colorectal Cancer: A Mendelian Randomisation Analysis." *Nature Communications* 11 (1): 597.

Pashos, C., Botteman, M., Laskin, B., and Redaelli, A. 2002. "Bladder Cancer: Epidemiology, Diagnosis, and Management." *Cancer Practice* 10 (6): 311–22.

Pedroso, T., Benvindo-Souza, M., Araújo Nascimento, F., Woch, J., Gonçalves Dos Reis, F., and Melo e Silva, M. 2022. "Cancer and Occupational Exposure to Pesticides: A Bibliometric Study of the Past 10 Years." *Environmental Science and Pollution Research*, 1–12.

Peltomäki, P., Olkinuora, A., and Nieminen, T. 2020. "Updates in the Field of Hereditary Nonpolyposis Colorectal Cancer." *Expert Review of Gastroenterology & Hepatology* 14 (8): 707–20. https://doi.org/10.1080/17474124.2020.1782187.

Pelucchi, C., Galeone, C., Tramacere, I., Bagnardi, V., Negri, E., Islami, F., et al. 2012. "Alcohol Drinking and Bladder Cancer Risk: A Meta-Analysis." *Annals of Oncology* 23 (6): 1586–93.

Perry, P.E., Thomson, E.J., Day, N.E., and Bartsch, H. 1983. "Induction of SCE by Opium Pyrolysates in CHO Cells and Human Peripheral Blood Lymphocytes." *Carcinogenesis* 4 (2): 227–30.

Edward , P., Luckett, B., Applebaum, K., Marsit, C., McClean, M., and Kelsey, K. 2008. "Dairy Products, Leanness, and Head and Neck Squamous Cell Carcinoma." *Head & Neck: Journal for the Sciences and Specialties of the Head and Neck* 30 (9): 1193–1205.

Platek, A., Cannioto, R., Etter, J., Kim, J., Joseph, J., Gulati, N., et al. 2017. "The Association of Lifetime Physical Inactivity with Head and Neck Cancer: A Hospital-Based Case–Control Analysis." *European Archives of Oto-Rhino-Laryngology* 274: 3773–80.

Pourshams, A, Khademi, H., Fazeltabar Malekshah, A., Islami, F., Nouraei, M., Sadjadi, A., et al. 2010. "Cohort Profile: The Golestan Cohort Study—a Prospective Study of Oesophageal Cancer in Northern Iran." *International Journal of Epidemiology* 39 (1): 52–59.

Poustchi, H., Eghtesad, S., Kamangar, F., Etemadi, A., Keshtkar, A., Hekmatdoost, A., et al. 2018. "Prospective Epidemiological Research Studies in Iran (the PERSIAN Cohort Study): Rationale, Objectives, and Design." *American Journal of Epidemiology* 187 (4): 647–55. https://doi.org/10.1093/aje/kwx314.

Pukkala, E. 1995. *Cancer Risk by Social Class and Occupation: A Survey of 109,000 Cancer Cases among Finns of Working Age. Contributions to Epidemiology and Biostatistics, Vol. 7*. Karger Medical and Scientific Publishers.

Pukkala, E., Guo, J., Kyyrönen, P., Lindbohm, M., Sallmén, M., and Kauppinen, T. 2005. "National Job-Exposure Matrix in Analyses of Census-Based Estimates of Occupational Cancer Risk." *Scandinavian Journal of Work, Environment & Health* 31 (2): 97–107.

Pukkala, E., Martinsen, J., Lynge, E., Kolbrun-Gunnarsdottir, H., Sparén, P., Tryggvadottir, L., et al. 2009. "Occupation and Cancer–Follow-up of 15 Million People in Five Nordic Countries." *Acta Oncologica* 48 (5): 646–790.

Rafiemanesh, H., Lotfi, Z., Bakhtazad, S., Ghoncheh, M., and Salehiniya, H. 2018. "The Epidemiological and Histological Trend of Bladder Cancer in Iran." *Journal of Cancer Research and Therapeutics* 14 (3): 532–36.

Ramström, L, and Wikmans, T. 2014. "Mortality Attributable to Tobacco among Men in Sweden and Other European Countries: An Analysis of Data in a WHO Report." *Tobacco Induced Diseases* 12: 1–4.

Rashidian, H., Hadji, M., Marzban, M., Gholipour, M., Rahimi-Movaghar, A., Kamangar, F., et al. 2017. "Sensitivity of Self-Reported Opioid Use in Case-Control Studies: Healthy Individuals versus Hospitalized Patients." *PloS One* 12 (8): e0183017. https://doi.org/10.1371/journal.pone.0183017.

Rashidian, H., Haghdoost, A.A., Hadji, M., Marzban, M., Gholipour, M., and Zendehdel, K. 2021. "Association between Opium Use and Bladder Cancer: A Case-Control Study in a High Risk Area of Iran." *Clinical Epidemiology and Global Health* 11: 100772.

Rawla, P, Sunkara, T., and Barsouk, A. 2019. "Epidemiology of Colorectal Cancer: Incidence, Mortality, Survival, and Risk Factors." *Gastroenterology Review/Przegląd Gastroenterologiczny* 14 (2): 89–103.

Ray, R., Kattimani, S., and Sharma, H.K. 2006. "Opium Abuse and Its Management: Global Scenario." *World Health Organization Department of Mental Health and Substance Abuse Management of Substance Abuse. National Drug Dependence Treatment Centre All India Institute of Medical Sciences New Delhi, India*, 1–13.

Rink, M., Crivelli, J., Shariat, S., Chun, F., Messing, E., and Soloway, M. 2015. "Smoking and Bladder Cancer: A Systematic Review of Risk and Outcomes." *European Urology Focus* 1 (1): 17–27.

Robsahm, T., Aagnes, B., Hjartåker, A., Langseth, H., Bray, F., and Larsen, I. 2013. "Body Mass Index, Physical Activity, and Colorectal Cancer by Anatomical Subsites." *European Journal of Cancer Prevention* 22 (6): 492–505.

Roshandel, G., Ghanbari-Motlagh, A., Partovipour, E., Salavati, F., Hasanpour-Heidari, S., Mohammadi, G., et al. 2019. "Cancer Incidence in Iran in 2014: Results of the Iranian National Population-Based Cancer Registry." *Cancer Epidemiology* 61 (2019): 50–58. https://doi.org/10.1016/j.canep.2019.05.009.

Sadeghi, A., Behmard, S., and Vesselinovitch, S. 1979. "Opium: A Potential Urinary Bladder Carcinogen in Man." *Cancer* 43 (6): 2315–21.

Sadjadi, A., Derakhshan, M., Yazdanbod, A., Boreiri, M., Parsaeian, M., Babaei, M., et al. 2014. "Neglected Role of Hookah and Opium in Gastric Carcinogenesis: A Cohort Study on Risk Factors and Attributable Fractions." *International Journal of Cancer* 134 (1): 181–88.

Saginala, K., Barsouk, A., Sukumar Aluru, J., Rawla, P., Padala, S.A, and Barsouk, A. 2020. "Epidemiology of Bladder Cancer." *Medical Sciences* 8 (1): 15.

Sanford, N., Giovannucci, E., Ahn, C., Dee, E., and Mahal, B. 2020. "Obesity and Younger versus Older Onset Colorectal Cancer in the United States, 1998–2017." *Journal of Gastrointestinal Oncology* 11 (1): 121.

Sanjosé, S., Serrano, B., Tous, S., Alejo, M., Lloveras, B., Quirós, B., et al. 2018. "Burden of Human Papillomavirus (HPV)-Related Cancers Attributable to HPVs 6/11/16/18/31/33/45/52 and 58." *JNCI Cancer Spectrum* 2 (4): pky045.

Sanli, O., Dobruch, J., Knowles, M., Burger, M., Alemozaffar, M., Nielsen, M., et al. 2017. "Bladder Cancer." *Nature Reviews Disease Primers* 3 (1): 1–19.

Santos, M., dos, F., Fernandes, G., Antunes, J., Villa, L., and Toporcov, T. 2021. "Global Incidence Trends in Head and Neck Cancer for HPV-Related and-Unrelated Subsites: A Systematic Review of Population-Based Studies." *Oral Oncology* 115: 105177.

Sapkota, A., Hsu, C.C., Zaridze, D., Shangina, O., Szeszenia-Dabrowska, N., Mates, D., et al. 2008. "Dietary Risk Factors for Squamous Cell Carcinoma of the Upper Aerodigestive Tract in Central and Eastern Europe." *Cancer Causes & Control* 19: 1161–70.

Sawicki, T., Ruszkowska, M., Danielewicz, A., Niedźwiedzka, E., Ar\lukowicz, T., and Przyby\lowicz, K. 2021. "A Review of Colorectal Cancer in Terms of Epidemiology, Risk Factors, Development, Symptoms and Diagnosis." *Cancers* 13 (9): 2025.

Schulz, K., and Grimes, D. 2002. "Case-Control Studies: Research in Reverse." *The Lancet* 359 (9304): 431–34.

Sciannameo, V., Carta, A., d'Errico, A., Giraudo, M., Fasanelli, F., Arici, C., et al. 2019. "New Insights on Occupational Exposure and Bladder Cancer Risk: A Pooled Analysis of Two Italian Case–Control Studies." *International Archives of Occupational and Environmental Health* 92: 347–59.

Shakeri, R, Kamangar, F., Mohamadnejad, M., Tabrizi, R., Zamani, F., Mohamadkhani, A., et al. 2016. "Opium Use, Cigarette Smoking, and Alcohol Consumption in Relation to Pancreatic Cancer." *Medicine* 95 (28).

Shakeri, R., Kamangar, F., Nasrollahzadeh, D., Nouraie, M., Khademi, H., Etemadi, A., et al. 2012. "Is Opium a Real Risk Factor for Esophageal Cancer or Just a Methodological Artifact? Hospital and Neighborhood Controls in Case-Control Studies." *PloS One* 7 (3): e32711.

Shakeri, R, Malekzadeh, R., Etemadi, A., Nasrollahzadeh, D., Aghcheli, K., Sotoudeh, M., et al. 2013. "Opium: An Emerging Risk Factor for Gastric Adenocarcinoma." *International Journal of Cancer* 133 (2): 455–61.

Shakhssalim, N., Hosseini, S.Y., Basiri, A., Eshrati, B., Mazaheri, M., and Soleimanirahbar, A. 2010. "Prominent Bladder Cancer Risk Factors in Iran." *Asian Pacific Journal of Cancer Prevention* 11 (3): 601–6.

Sheikh, M., Kamangar, F., and Malekzadeh, R. 2020. "Fifty Years of Research and One Conclusion: Opium Causes Cancer." *Archives of Iranian Medicine* 23 (11): 757–60.

Sheikh, M., Shakeri, R., Poustchi, H., Pourshams, A., Etemadi, A., Islami, F., et al. 2020. "Opium Use and Subsequent Incidence of Cancer: Results from the Golestan Cohort Study." *The Lancet Global Health* 8 (5): e649–60. https://doi.org/10.1016/S2214-109X(20)30059-0.

Shoffel-Havakuk, H., Cohen, O. Slavin, M., Haimovich, Y., Halperin, D., and Lahav, Y.. 2018. "Intravenous Opioid Drug Abuse as an Independent Risk Factor for Supraglottic Squamous Cell Carcinoma—A Case-Control Study." *Clinical Otolaryngology* 43 (2): 456–62.

Shuster, J., and Cook, B. 1983. "Hospital or Population Controls: A Discussion." *Journal of Chronic Diseases* 36 (4): 315–16.

Siegel, R., Miller, K., and Jemal, A. 2019. "Cancer Statistics, 2019." *CA: A Cancer Journal for Clinicians* 69 (1): 7–34.

Simon, K. 2016. "Colorectal Cancer Development and Advances in Screening." *Clinical Interventions in Aging*, 967–76.

Singh, G., Jaiswal, A., Goel, A., and Raghav, P. 2021. "Opium Usage and Risk of Head and Neck Cancer: A Systematic Review and Meta-Analysis." *Asian Pacific Journal of Cancer Prevention* 22 (3): 661.

Slattery, M.L., Levin, T.R., Ma, K., Goldgar, D., Holubkov, R., and Edwards, S. 2003. "Family History and Colorectal Cancer: Predictors of Risk." *Cancer Causes & Control* 14: 879–87.

Slattery, M., and Kerber, R. 1994. "Family History of Cancer and Colon Cancer Risk: The Utah Population Database." *JNCI: Journal of the National Cancer Institute* 86 (21): 1618–26.

Sohrabi, M., Gholami, A., Hosseini Azar, M.H, Yaghoobi, M., Mirasgari Shahi, M., Shirmardi, S., et al. 2018. "Trace Element and Heavy Metal Levels in Colorectal Cancer: Comparison between Cancerous and Non-Cancerous Tissues." *Biological Trace Element Research* 183 (1): 1–8.

Sormunen, J., Talibov, M., Martinsen, J., Kjaerheim, K., Sparen, P., Tryggvadottir, L., et al. 2016. "Perceived Physical Strain at Work and Incidence of Colorectal Cancer: A Nested Case–Control Study." *Cancer Epidemiology* 43: 100–104.

Stanford-Moore, G., Bradshaw, P., Weissler, M., Zevallos, J., Brennan, P., Anantharaman, D., et al. 2018. "Interaction between Known Risk Factors for Head and Neck Cancer and Socioeconomic Status: The Carolina Head and Neck Cancer Study." *Cancer Causes & Control* 29: 863–73.

Sundbøll, J., Thygesen, S., Veres, K., Liao, D., Zhao, J., Gregersen, H., et al. 2019. "Risk of Cancer in Patients with Constipation." *Clinical Epidemiology* 11 (2019): 299–310. https://doi.org/10.2147/CLEP.S205957.

Sung, H., Ferlay, J., Siegel, R., Laversanne, M., Soerjomataram, I., Jemal, A., et al. 2021. "Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries." *CA: A Cancer Journal for Clinicians* 71 (3): 209–49. https://doi.org/10.3322/caac.21660.

Swanson, G., Belle, S.H., and Burrows-Jr, R.W.. 1985. "Colon Cancer Incidence among Modelmakers and Patternmakers in the Automobile Manufacturing Industry: A Continuing Dilemma." *Journal of Occupational Medicine*, 567–69.

Taziki, M.H., Fazel, A., Salamat, F., Sedaghat, S.M., Ashaari, M., Poustchi, H., et al. 2018. "Epidemiology of Head and Neck Cancers in Northern Iran: A 10-Year Trend Study from Golestan Province." *Archives of Iranian Medicine* 21 (9): 406–11.

Tourangeau, R., and Yan, T. 2007. "Sensitive Questions in Surveys." *Psychological Bulletin* 133 (5): 859.

Toutounchi, M., Mazdak, H., Najafipour, S.H., and Soleymani, B. 2000. "Bladder Cancer Risk Factors among Isfahan Population: A Case-Control Study." *Journal of Research in Medical Science* 5: 151–56.

"U.S. National Institutes of Health, National Cancer Institute." 2023. https://www.cancer.gov/types/head-and-neck/head-neck-fact-sheet.

Ubago-Guisado, E., Rodríguez-Barranco, M., Ching-López, A., Petrova, D., Molina-Montes, E., Amiano, P., et al. 2021. "Evidence Update on the Relationship between Diet and the Most Common

Cancers from the European Prospective Investigation into Cancer and Nutrition (EPIC) Study: A Systematic Review." *Nutrients* 13 (10): 3582.

Vallejo, R., Leon-Casasola, O., and Benyamin, R. 2004. "Opioid Therapy and Immunosuppression: A Review." *American Journal of Therapeutics* 11 (5): 354–65.

VanderWeele, T. 2015. *Explanation in Causal Inference: Methods for Mediation and Interaction*. New York, NY: Oxford University Press.

VanderWeele, T, and Vansteelandt, S. 2014. "Mediation Analysis with Multiple Mediators." *Epidemiologic Methods* 2 (1): 95–115.

Walboomers, J., Jacobs, M., Manos, M., Bosch, F., Kummer, J., Shah, K., et al. 1999. "Human Papillomavirus Is a Necessary Cause of Invasive Cervical Cancer Worldwide." *The Journal of Pathology* 189 (1): 12–19.

Wang, C., and Jiang, H. 2012. "Meat Intake and Risk of Bladder Cancer: A Meta-Analysis." *Medical Oncology* 29: 848–55.

Warnakulasuriya, S., Cronin-Fenton, D., Jinot, J., Kamangar, F., Malekzadeh, R., Dar, N.A., et al. 2020. "Carcinogenicity of Opium Consumption." ELSEVIER SCIENCE INC STE 800, 230 PARK AVE, NEW YORK, NY 10169 USA. https://www.iarc.who.int/infographics/opium-consumption.

WHO. 2012. "WHO Global Report on Mortality Attributable to Tobacco," *https://www.who.int/* (March 2023).

United Nations Office on Crime. 2019. "World Drug Report 2019." *https://wdr.unodc.org/wdr2019/*.

Wu, Z., Han, Y., Caporaso, J., Bokulich, N., Mohamadkhani, A., Moayyedkazemi, A., et al. 2021. "Cigarette Smoking and Opium Use in Relation to the Oral Microbiota in Iran." *Microbiology Spectrum* 9 (2): e00138-21. https://doi.org/10.1128/Spectrum.00138-21.

Yaksh, T., and Wallace, M. 2011. "Opioids, Analgesia, and Pain Management." *Goodman and Gilman's the Pharmacological Basis of Therapeutics*, 481–526.

Ye, P., Xi, Y., Huang, Z., and Xu, P. 2020. "Linking Obesity with Colorectal Cancer: Epidemiology and Mechanistic Insights." *Cancers* 12 (6): 1408.

Zeighami, S., Azizzadeh, E., Tabatabaee, H.R., Adib, A., Babaei, A.H., and Ariafar, A. 2018. "Opium and Grade of Urothelial Bladder Cancer." *Journal of Nephropathology* 7 (2).

Zeppetella, G. 2011. "Opioids for the Management of Breakthrough Cancer Pain in Adults: A Systematic Review Undertaken as Part of an EPCRC Opioid Guidelines Project." *Palliative Medicine* 25 (5): 516–24.

Zhao, J., Stockwell, T., and Macdonald, S. 2009. "Non–Response Bias in Alcohol and Drug Population Surveys." *Drug and Alcohol Review* 28 (6): 648–57.

PUBLICATION I

The Iranian Study of Opium and Cancer (IROPICAN): Rationale, Design, and Initial findings

Maryam Hadji, Hamideh Rashidian, Maryam Marzban, Mahin Gholipour, Ahmad Naghibzadeh-Tahami, Elham Mohebbi, Elmira Ebrahimi, Bayan Hosseini, AliAkbar Haghdoost, Abbas Rezaianzadeh, Afarin Rahimi-Movaghar, Abdolvahab Moradi, Monireh Sadat Seyyedsalehi, Reza Shirkoohi, Hossein Poustchi, Sareh Eghtesad, Farid Najafi, Roya Safari-Faramani, Reza Alizadeh-Navaei, Ali Reza Ansari Moghadam, Mahdieh Bakhshi, Azim Nejatizadeh, Masumeh Mahmudi, Soudabeh Shahid-Sales, Saideh Ahmadi-Simab, Omid Nabavian, Paolo Boffetta, Eero Pukkala, Elisabete Weiderpass, Farin Kamangar, Kazem Zendehdel

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Original Article

The Iranian Study of Opium and Cancer (IROPICAN): Rationale, Design, and Initial Findings

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Abstract

Background: The International Agency for Research on Cancer (IARC) recently classified opium use as a Group 1 carcinogen. However, much remains to be studied on the relation between opium and cancer. We designed the Iranian Opium and Cancer (IROPICAN) study to further investigate the association of opium use and cancers of the head and neck, bladder, lung, and colon and rectum. In this paper, we describe the rationale, design, and some initial results of the IROPICAN Study.

Methods: The IROPICAN is a multi-center case-control study conducted in 10 provinces of Iran. The cases were all histologically confirmed and the controls were selected from hospital visitors who were free of cancer, were not family members or friends of the cancer patients, and were visiting the hospital for reasons other than their own ailment. The questionnaires included detailed questions on opium use (including age at initiation, duration, frequency, typical amount, and route), and potential confounders, such as tobacco use (e.g., cigarettes, *nass* and water-pipe), and dietary factors. Biological samples, including blood and saliva, were also collected.

Results: The validation and pilot phases showed reasonably good validity, with sensitivities of 70% and 69% for the cases and controls, respectively, in reporting opium use. The results also showed excellent reliability, with intra-class correlation coefficients of 0.96 for ever opium use and 0.88 (95% CI: 0.80, 0.92) for regular opium use. In the main phase, we recruited 3299 cancer cases (99% response rate) and 3477 hospital visitor controls (89% response rate). The proportion of ever-use of opium was 40% among cases and 18% among controls.

Conclusion: The IROPICAN study will serve as a major resource in studies addressing the effect of opium on risk of cancers of the head and neck, bladder, lung, and colon and rectum.

Keywords: Bladder cancer, Colorectal cancer, Head and neck cancer, Lung cancer, Opium, Protocol

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Introduction

Opium, a highly addictive substance obtained from the unripe seedpod of the poppy plant, is illicitly consumed by millions of people around the world, particularly in central Asian countries.[1] Freshly taken from the poppy plant, opium contains alkaloids (e.g., morphine, codeine, and thebaine) and non-alkaloids (e.g., water, sugars, fat, and meconic acid). It is often minimally processed by heating, boiling, and drying, and variably adulterated with some chemicals (e.g., lead or chromium) before it reaches consumers. In the minimally processed form, opium may be consumed as raw opium (*teriak*), opium sap (*shireh*), or opium dross (*sukhteh*).[2] These forms of opium may be ingested or smoked. Therefore, similar to tobacco, opium is a complex substance with many chemicals.

Case-control and cohort studies, conducted mainly within the past two decades, have provided substantial evidence that opium use could increase the risk of overall death, cardiovascular mortality,[3] and cancer mortality.[4,5] Opium use has been found to be associated with higher risk of cancers of the larynx,[4-7] bladder,[8-10] lung,[10-12] pharynx,[6,7,13,14] stomach,[10,15,16] esophagus,[10,17-20] pancreas[10,21,22] and colon and rectum.[23,24] Using data from these studies, complemented by mechanistic studies, the International Agency for Research on Cancer (IARC) recently classified opium use a Group 1 carcinogen, i.e. carcinogenic in humans.[25,26]

The IARC Working Group concluded that opium use causes cancers of the larynx, lung, and bladder. However, there remain substantial knowledge gaps for the association of opium use and cancer. For example, elucidating the magnitude of the association for these cancers and studying the interaction of opium use with tobacco smoking for these cancers need further investigation. Despite the availability of some credible data, it is unclear whether opium causes cancers of the esophagus, stomach, or pancreas. Data for some cancers, most notably those of colon and rectum, are contradictory.[10,24] While epidemiologic findings so far suggest that consumption of opium in its various forms (*teriak*, *shireh* and *sukhteh*) and major exposure routes (ingestion and smoking) would be carcinogenic, there is a clear need for further data to support these findings. To advance the science in this field, we initiated the Iranian Opium and Cancer (IROPICAN) Study in 2012, to evaluate the association of opium exposure with risk of cancers of the head and neck, lung, bladder, and colon and rectum.

We aimed to advance the field by planning studies that had a substantially larger sample size than any studies before, and to collect detailed data on opium use needed for dose-response analyses, controlling for potential confounders, and evaluation of possible reverse causality.[27] The findings for head and neck cancer are already published which showed odds ratio (OR) (3.76, 95% CI: 2.96–4.79) for regular opium use[6] and contributed to the recent IARC Working Group conclusions, particularly for laryngeal and pharyngeal cancer evaluation. The data for the remaining cancer sites are being analyzed. While the IROPICAN study is primarily focused on opium use, the researchers have collected substantial data on other important exposures (e.g., waterpipe use) and collected biological samples that can be used for a host of epidemiological studies. Therefore, a clear and thorough description of methods and available data will be of interest for the readers of future studies.

The objective of this paper is to introduce the

IROPICAN study, including the study setting, design of the questionnaires, validity and reliability of the responses, biological samples collected during the study, the rationale for selection of controls, and the potential for in-depth evaluation of cancer risk factors using the material collected in these studies.

Materials and Methods

Study Setting

The IROPICAN case-control study was conducted in Iran, a country with a high consumption rate of opium[1] that has a strong infrastructure for conducting large-scale studies. It was a multi-center study, conducted in 10 provinces that were estimated to have medium to high opium use,[28] namely Tehran, Kerman, Fars, Golestan, Khorasan-Razavi, Kermanshah, Mazandaran, Bushehr, Hormozgan, and Sistan-Baluchestan (Figure 1), to evaluate the association between opium consumption and risk of cancers of the head and neck, lung, bladder, and colon and rectum. The regions were selected based on prevalence of opium use and availability of cancer care centers for recruiting patients and contols. Of these, Tehran, Kerman, Fars, and Golestan were the first provices to join the study in 2014 to 2015. Later, Kemanshah, Mazandaran, and Khorasan-Razavi joined in 2017 and 2018, followed by Hormozgan, Sistan-Blouchestan, and Bushehr in 2018 to 2020.

Questionnaires

We developed a detailed questionnaire and administered it to all study participants. This questionnaire was adapted from the Golestan Cohort Study,[29] because it had detailed data on opium use, had been tested extensively in the field (in the cohort and several case-control studies), was the basis of many publications, and could be eventually used for combined analyses of data. The IROPICAN questionnaire include detailed questions on opium use, including types of opium used (e.g. *teriak*, *shireh*, *sukhteh*), routes of opium use (smoking versus ingestion), age of initiating and stopping opium use, and frequency and amount of opium use. The questionnaire also include extensive data on covariates, such as: demographic and socioeconomic factors (e.g. age, ethnicity, rural/urban status, education, occupational history, various amenities owned; physical activity (using the international physical activity questionnaire, IPAQ[30]); information on various aspects of health (oral health, female reproductive history, history of various diseases, personal and family history of cancer); life-long consumption of tobacco in the forms of cigarettes, water-pipe, pipe, *naswar*, and *chopogh* (a special pipe); history of alcohol consumption; and history of opioid use (e.g. diphenoxylate, methadone, morphine, codeine); history of other illicit drug use (e.g. heroin, heroin crack, hashish, tramadol, methadone). The crack used in Iran is heroin-based and different from the cocaine-based[34] crack used in Western countries. We used a validated and reproducible 130-item food frequency questionnaire similar to that used in the Persian Cohort Study, a large study aiming to recruit 150 000 Iranians aged 35–70 years from 18 regions of Iran.[31]

Figure 1. Distribution of the Iran Opium and Cancer (IROPICAN) study sites in different provinces of Iran.

Physical Examination and Biological Sample Collection

All prospective study participants underwent physical examination using a standard protocol and performed by trained personnel. Weight was measured using a portable non-digital weight scale (Fazeni), height with a height scale (Seca), and blood pressure was measured with a digital scale (Haalar).[32,33]

Blood samples were collected to study the presence of different opium adulterants[34] and their associations with selected cancers. We collected 12 mL of venous whole blood for each cancer case and control. From this sample, 6 mL is kept in a vacutainer tube with EDTA anticoagulant for DNA extraction; 3 mL is stored for RNA extraction, and 3 mL is stored in a vacutainer tube without anticoagulant for serum extraction. One drop of blood is stored in a DNA banking card. Oral saliva from head and neck cancer cases and their controls is collected for assessing the prevalence of human papillomavirus (HPV) and its serotypes.

Blood and saliva samples were stored at minus 20 degrees Celsius in each province and shipped to the main center every month (maximum two months after collection) for storage at minus 80 degrees Celsius. DNA extraction was done using the salting-out method at the Cancer Research Center (CRC), Cancer Institute of Iran; extracted DNAs were stored at minus 20 degrees Celsius. These biological samples will be used for several other projects in the future including potential genome-wide association studies in international consortia.

Interviewer Training and Project Management

Interviewers with a bachelor's or master's degree in public health or biology were trained intensively to use a standard protocol, to carry out standardized interviews, and to collect samples for the study. One experienced researcher in each province was responsible for the management of field-work and data collection. In addition, a central coordinator based in the main center (Cancer Research Center in Tehran University) tracked all implemented work daily, and made a visit to each center twice a year to supervisor the operations. Each center regularly reported the number of cases and controls recruited.

Choosing the Most Appropriate Control Group Using a Validation Study

Self-reported opium use in the Golesan Cohort Study was strongly correlated with urine morphine metabolites in a sample of 130 study cohort participants[35]; using urine codeine or morphine as the gold standard for use of opium, self-report had a sensitivity of 0.93 and a specificity of 0.89. However, the validity of this questionnaire outside the Golestan Province was unclear. Therefore, in June-July 2016, a validation study was conducted in four provinces that were first selected as study sites – Kerman, Fars, Golestan, and Tehran – to estimate the magnitude of underreporting and to determine the best potential control group.

The two potential control choices were: 1) hospitalized non-cancer patients (178 men); and 2) healthy hospital visitors who were companions of patients with chronic diseases (186 men). The prevalence of self-reported regular opioid use, defined as using at least once a week for six months, was 36% (95% confidence interval: 28%, 43%) in hospitalized patients versus 19% (95% CI: 13%, 25%) in healthy individuals.[36] Compared to a gold standard of using urine rapid drug screen and thin layer chromatography, self-reported use had a sensitivity of 77% in hospitalized patients and 69% in healthy visitors (P = 0.4).[36] After correction for sensitivity, the frequency of regular opioid use (primarily opium use) was 47% and 28% in hospitalized patients and healthy visitors individuals, respectively. Because 47% was a substantial overestimation of the estimated prevalence of use in the population, and that sensitivity of self-reported use was comparable in the two control groups, we opted for the healthy visitor controls in the IROPICAN Study.

Validation of Self-reported Opium Use in Cases

We studied the sensitivity of self-reported opium use in a study of 100 cancer patients (with various types of cancer) who were slated for surgery in the Cancer Institute of Tehran University of Medical Sciences. We recruited only men, to make the results comparable with the validation study in controls, with mean age of 61.5 (standard deviation, SD: 16.3) years. Most cancer patients had used opioids to alleviate pain; therefore, urine test results could not be used to validate responses. Instead, we used patient's response to the anesthesiologist's questions as gold standard; anesthesiologists routinely ask these questions before each surgery to prevent withdrawal syndrome. Sensitivity of self-reported opium use was 69.6%.

Reliability Study

The Golestan Cohort Study questionnaire had shown excellent reliability for ever opium use report, with a kappa of 0.96.[35] To confirm again, we assessed the reliability of the questions of drug, tobacco, and alcohol use in 57 patients (47 men and 10 women) who were referred to an addiction control center. We used a test-retest approach, with the second administration done two weeks after the first. The intraclass correlation coefficient (ICC) of lifetime opium use was 0.96, and the ICC for regular use of opium was 0.88 (95% CI, 0.80, 0.92). The ICC was 0.78 (95% CI: 0.63, 0.90) for method of opium use and 0.97 (95% CI: 0.95, 0.98) for opium type (Table 1).

Pilot Study

In the pilot study conducted in January-April 2017, we recruited patients with four types of cancer as the case group and healthy visitors as the control group from referral cancer care centers and clinics in the four provinces

Table 1. Intra-class Correlation (ICC) and 95% Confidence Interval (CI) for Questions on Opium, Other Drugs, Tobacco and Alcohol Use in the Iranian Opium and Cancer (IROPICAN) Multi-center Case-Control Study, 2015–2020

Variables	ICC	95% CI
Ever cigarette use	0.96	(0.94, 0.97)
Regular cigarette use	0.95	(0.91, 0.97)
Ever water pipe use	0.88	(0.80, 0.93)
Regular water pipe use	0.99	(0.98,0.99)
Ever *chopogh* use	0.88	(0.80, 0.93)
Regular *chopogh* use	0.96	(0.93, 0.98)
Ever opium use	0.96	(0.93, 0.96)
Regular opium use	0.88	(0.80, 0.92)
Ever heroin use	0.95	(0.92, 0.97)
Regular heroin use	0.92	(0.86, 0.95)
Ever hashish use	0.98	(0.96, 0.98)
Regular hashish use	0.91	(0.85, 0.95)
Ever codeine use	0.89	(0.81, 0.93)
Regular codeine use	0.94	(0.89, 0.96)
Ever tramadol use	0.90	(0.82, 0.94)
Regular tramadol use	0.84	(0.72, 0.91)
Ever diphenoxylate use	0.95	(0.92, 0.97)
Regular diphenoxylate use	0.84	(0.71, 0.91)
Ever methadone use	0.94	(0.90, 0.96)
Regular methadone use	0.97	(0.96, 0.98)
Ever alcohol use	0.98	(0.96, 0.98)
Regular alcohol use	0.99	(0.98, 0.99)
Type of opium	0.97	(0.95, 0.98)
Route of opium use	0.78	(0.63, 0.90)

that were initially selected as study sites (Tehran, Shiraz, Kerman, and Golestan). The objectives of the pilot study were to: (*a*) determine the feasibility of the case-control study; (*b*) optimize the questionnaires; (*c*) train partnering research institutions for data collection; (*d*) assess the geographic variation of opium use and other opiates in different regions of Iran; and (*e*) optimize biological sample collection methods. The pilot study showed that conducting the study was feasible. Based on pilot study findings, we made minor revisions to the questionnaire, primarily focused on questions regarding opium use and other drug use. For example, a question about frequency of opium use per day/week/month/year was added. The other aims of the pilot study were also achieved. Only minor changes were made in sample collection and methodology when the full project was set up. Partnering institutions received the required training. Also, variability of the opium consumption was determined. Following a successful pilot study, we calculated the required sample size and started recruitment of the cases and controls.

Sample Size Calculation

While it is difficult to accurately estimate the prevalence of opium use as an illicit substance[37] the use of which is also associated with some social stigma,[38] the results of national surveys and our pilot results suggested that a prevalence of approximately 20% among the controls was a reasonable assumption. Assuming a prevalence of 20% of opium use in the Iranian population[39,40] living in high prevalence regions, 90% power and a two-sided significance level of 0.05, we estimated that 250 cases and 250 controls would be sufficient to detect an odds ratio of 2.0. However, we chose to aim at a sample size of 800 cases for each type of cancer (i.e. 3200 cancer cases overall) and 3200 common controls, to allow for various subgroup analyses.[39,40] Where possible, we exceeded this number. As of the date this report, we included 743 lung cancers, 920 colorectal cancers, 717 bladder cancers, and 919 head and neck cancers.

Case Recruitment

Patients with confirmed histological primary cancers of the lung (ICD-10: C33-34), colorectum (C18-21), bladder (C67), or squamous cell carcinoma (SCC) of the head and neck (i.e., oral cavity (C03-06), larynx (C32) and pharynx (C10-C14)) admitted to referral hospitals of the universities of medical sciences of 10 provinces (Tehran, Kerman, Fars, Golestan, Khorasan-Razavi, Kermanshah, Mazandaran, Bushehr, Hormozgan, and Sistan-Baluchestan), were recruited as cases. We only included newly diagnosed resident cancer cases, defined as those who had received a diagnosis within one year prior to the date of interview and who had resided at least two years in the study region. Other inclusion criteria were being of Iranian nationality, ability to speak and understand the Farsi language, ability to participate in an interview taking about 80 minutes, and being in the age range of 30 to 75 years. Patients with metastatic cancers, second primary cancers, and those without a confirmed pathology report were excluded. Pregnant and nursing women were also excluded from the study.

Control Recruitment

The control study participants (hospital visitors) were enrolled concurrently with the cases. The controls (who had to be free of cancer) were chosen from the relatives or friends of patients from non-oncology wards, or others who visited the hospital for reasons other than receiving treatment. Choosing healthy visitors was a conclusion that we had reached during the validation studies (see above). Another rationale for this choice of controls was that we aimed at excluding those who were ill (hence, more likely than the general population to use opium) and family members or friends of patients with cancer (who are likely to share many habits, including opium use) with the cases. Other inclusion criteria were similar to those of cases, i.e., being of Iranian nationality, ability to speak and understand the Farsi (Persian) language, ability to participate in an interview taking about 80 minutes, and being in the age range of 30 to 75 years. We aimed at enrolling controls who were frequency-matched to the cases for gender, age (five year intervals), and place of residence (by province and capital city/non-capital city).

Data Processing and Exposure Assessment

Ever-use of opium was defined as using opium at least once during lifetime and regular opium use was defined as using opium at least once a week for at least six consecutive months. The cumulative amount of lifetime opium use was defined as the amount of opium use (grams per day) multiplied by days of opium use during lifetime.

We calculated pack-years, starting age and stopping age of cigarette use to be used as the main measure of exposure to cigarette smoking. Other types of tobacco used in the area are water pipe, pipe, and *chopogh* (a special type of pipe with a long tube).[41] For water pipe, the head-year unit was calculated for cumulative use.[45] For estimation of use of alcohol, one standardized unit was defined as follows: 340 cc of beer; 150 cc of wine or champagne; or 40 cc of vodka, fortified wine or whisky. We provided an album containing pictures of standard units such as pilsner, glass, cocktail and tablespoon to participants to report their portion size according to the standardized units.

We created a composite variable for socioeconomic status using a host of variables, as described elsewhere.[42]

Results

Overall, 3299 cancer patients (743 with lung cancer, 920 with colorectal cancer, 717 with bladder cancer and 919 with head or neck cancer) and 3477 controls participated

in the main study. The response rate was 99% (3299 out of 3314) among the cases who were capable to participate and 89% (3477 out of 3925) among the controls.

Non-participants

Only 1% of the invited cases and 11% of the invited controls chose not to participate in the study. The main stated reasons for non-participation were sickness and lethargy among cancer patients (93% of the non-participants) and lack of time or unwillingness to donate a biological sample among controls (83% of the non-participants). Among the 15 cases who refused to participate, the mean age was 58.8 (SD 11.2) years and 66% were males. Among controls who refused to participate, the mean age was 58.3 (SD 10.9) years and 71% were males.

Demographic and Geographic Characteristics of the Participants

Table 2 shows the demographic characteristics and geographic distribution of the study participants by case status. The mean age on the interview date was 59.9 (SD 11.9) years for cases and 57.3 (SD 11.6) years for controls.

Opium Use

In all, 39.8% of cases and 17.5% of controls reported ever use of opium. Patients may start using opium to alleviate their pain – a prodromal symptom of subsequently diagnosed cancer. Opium's alkaloid constituents, in addition to having narcotic effects, may be used for medicinal purposes. For example, morphine has strong analgesic effects and may be used to alleviate pain in cancer patients, or codeine may be used to suppress cough.[43,44] If cases and controls who started using opium less than 3 years before their interview date were classified as non-users, 35.1% of cases and 13.0% of controls were classified as opium users (Table 3).

The median of lifetime use of opium among users were 0.9 (25th and 75th percentiles 0.3 and 3.0) gram per day among cases and 0.4 (0.1, 2.0) gram per day among controls.

The types of opium consumed differed among opium users: 28.0% of cases and 11.8% of controls used *teriak* (Table 3). The most common opium consumption method was smoking alone (21.4% of cases, 10.9% of controls).

Tobacco Use

The prevalence of cigarette ever use was 51.4% and 33.4% among cases and controls, respectively. The proportion of regular cigarette smokers was 48.4% in cases and 28.1% in controls. The prevalence of regular water-pipe use among cases and controls was 9.1% and 7.5%, respectively. The prevalence of regularly chewing *nass* was 1.7 among cases and 0.8 among controls (Table 3).

Biological Samples

Biological samples including whole blood, serum, plasma,

Table 2. Demographic Characteristics of Cases and Controls Enrolled in the Multi-center Case-Control Study of Opium and Cancer (IROPICAN Study), 2015-2020, by Cancer Site

Variable	Cases, Site No. (%)				Controls, No. (%)
	Lung	Colorectal	Bladder	Head and Neck	
Total	743 (100)	920 (100)	717 (100)	919 (100)	3477 (100)
Age					
<30	2 (0.3)	7 (0.8)	3 (0.4)	9 (1.0)	21 (0.6)
30–39	22 (3.0)	60 (6.5)	11 (1.5)	55 (6.0)	236 (6.8)
40–49	91 (12.3)	133 (14.5)	50 (7.0)	145 (15.8)	559 (16.1)
50–59	231 (31.1)	258 (28.0)	181 (25.2)	277 (30.1)	1070 (30.8)
60–69	257 (34.6)	274 (29.8)	267 (37.2)	295 (32.1)	1092 (31.4)
>70	138 (18.6)	187 (203)	205 (28.6)	138 (15.0)	499 (14.4)
Missing	2 (0.3)	1 (0.1)	-	-	-
Gender					
Female	181 (24.4)	387 (41.1)	93 (13.0)	232 (25.2)	1077 (31.0)
Male	562 (75.6)	533 (57.9)	624 (87.0)	687 (74.8)	2400 (69.0)
Province					
Tehran	148(19.9)	170 (18.5)	139 (19.4)	163 (17.7)	816 (23.5)
Fars	233 (31.4)	265 (28.8)	166 (23.2)	379 (41.2)	943 (27.1)
Kerman	128 (17.2)	112 (12.2)	150 (20.9)	160 (17.4)	525 (15.1)
Golestan	64 (8.6)	156 (17)	46 (6.4)	46 (5.0)	374 (10.8)
Mazandaran	36 (4.9)	59 (6.4)	24 (3.4)	17 (1.9)	136 (3.9)
Kermanshah	42 (5.7)	71 (7.7)	52 (7.3)	37 (4.0)	251 (7.2)
Khorasan-Razavi	17 (2.3)	87 (9.5)	30 (4.2)	44 (4.8)	170 (4.9)
Bushehr	38 (5.1)	*	56 (7.8)	**	84 (2.4)
Hormozgan	20 (2.3)	*	27 (3.8)	37 (4.0)	78 (1.9)
Sistan-Balouchestan	17 (2.3)	*	27 (3.8)	36 (3.9)	100 (2.9)

*Colorectal cancer cases were not enrolled in Bushehr, Hormozgan, and Sistan-Baluchestan.

**Head and neck cancer were not enrolled in Bushehr.

Table 3. Opium and Tobacco Use among the Cases and Controls in the Iranian Opium and Cancer (IROPICAN) Multi-center Case-Control Study of Opium and Cancer (IROPICAN Study), 2015–2020

Variable	Cases, No. (%)	Controls, No. (%)
Total	3299 (100)	3477 (100)
Opium use*		
Never used	1985 (60.2)	2870 (82.5)
Ever used	1314 (39.8)	607 (17. 5)
Non-regular use	129 (3.9)	139 (4.0)
Regular use	1185 (35.9)	468 (13.5)
Median opium amount		
Never used	1985 (60.1)	2870 (82.5)
Non-regular use	129 (3.9)	139 (0.4)
≤0.4 g/d	315 (10.0)	217 (6.2)
>0.4 g/d	870 (26.4)	251 (7.2)
Opium use duration		
Never used	1985 (60.1)	2870 (82.5)
Non-regular use	129 (3.9)	139 (4.0)
≤17 years	416 (12.6)	243 (7.0)
>17 years	769 (23.3)	225 (6.5)
Initiation age of opium use		
Never used	1985 (60.1)	2870 (82.5)
≤30 years	648 (19.6)	296 (8.5)
>30 years	577 (17.5)	262 (7.5)
Unknown	89 (2.7)	49 (1.4)
Type of opium use		
Never used	1985 (60.1)	2870 (82.5)
Non-regular use	129 (3.9)	139 (4.0)
Crude opium (*Teriak*)	922 (28.0)	411 (11.8)
Opium juice (*Shireh*)	89 (2.7)	30 (0.9)
Opium dross (*Sookhteh*)	3 (0.1)	2 (0.06)
More than one type	171 (5.2)	25 (0.7)
Route of opium use		
Never used	1985 (60.1)	2870 (82.5)
Non-regular use	129 (3.9)	139 (4.0)
Smoking	707 (21.4)	380 (10.9)
Ingestion	135 (4.1)	31 (0.9)
Inhalation	21 (0.6)	10 (0.3)
More than one route	318 (9.6)	45 (1.3)
Unknown	4 (0.1)	2 (0.06)
Cigarette use		
Never used	1602 (48.6)	2316 (66.6)
Non-regular use	101 (3.1)	184 (5.3)
Regular use	1596 (48.3)	977 (28.1)
Tobacco use		
Never used	2932 (88.9)	3177 (91.4)
Regular water pipe use	300 (9.1)	261 (7.5)
Regular *chopogh* use	2 (0.06)	1 (0.03)
Regular pipe use	9 (0.3)	9 (0.3)
Regular *nass*	56 (1.7)	29 (0.8)

extracted DNA for 2441 cases and 2768 controls, and saliva sample for 657 head and neck cancer cases, and 677 controls are available.

Discussion

The IROPICAN was designed and tested in various phases to serve as a high-quality study to evaluate the causal association of opium with various cancers. The results of the validation and pilot phases guided the investigators to choose the best potential control group, and to optimize the questionnaire, data collection, and biological sample collection methods. They also showed that the main study was feasible. The response rate was high in both cases and controls. Matching of controls for age, gender, province, and place of residence was feasible. However, the matching was not perfect, as the controls were slightly younger than the cases and they were more likely to be from Tehran and Fars provinces (Figure 2), two active centers that joined the study very early on.

The IROPICAN study has several advantages in evaluating the association of opium and cancers. This study has detailed information on opium use, including age of initiation; typical amount, frequency, duration of use; and types and routes of opium use. It also has data on potential important confounders, such as age, sex, tobacco use, and socioeconomic status. Since we have detailed data on amount of use over time, we will be able to examine dose-response associations. Reverse causality can be addressed in this study by removing those who initiated opium use within the three years prior to the interview. This is important, as some people with cancer may use opium to alleviate their cancer-related pain.[22] Indeed, the IROPICAN Study has already contributed to the evaluation of carcinogenicity of opium use.[6,25]

A major challenge in health studies on the effects of opium use is gathering reliable data on usage.[45] We believe that we are using a reasonably accurate method to collect data on opium use and other banned substances. We found the sensitivity of self-reporting of opium use at 77% and 69% among disease-controls and visitor-controls, respectively.[36] The high participation rates and the low denial of drug consumption in controls in our study may be due to the substantial training and supervision of our interviewers. It could also be due to the fact that opium use is quite common in Iran. So, although stigmatized, it is not quite as big a taboo as in some other countries. Fortunately, the sensitivity of reporting was similar in disease-controls and visitor-controls. This is in contrast to studies performed outside Iran that have reported substantial underreporting among controls.[46,47] In addition, we found a sensitivity of 70% for self-reporting opium use among cancer patients, which was similar to underreporting of healthy visitor controls. Although in case-control studies, information bias is typically higher in the controls than in the cases and

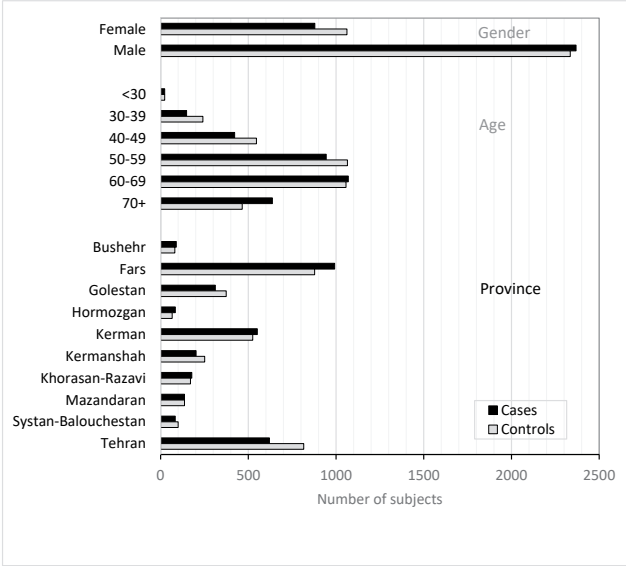

Figure 2. Numbers of Cases and Controls in the IROPICAN Study, by Gender, Age and Province.

leads to differential misclassification, here we found that underreporting for opium use was similar in the cancer patients and in the controls. Our findings suggest that we need to adjust for non-differential bias when we study the association between opium use and risk of cancers

In addition to accurate data on opium use, we have collected data on a host of other risk factors, such as various forms of tobacco use (e.g., cigarettes, water-pipe, and *nass*), which can be used for future epidemiologic studies. This wealth of data will also allow us to assess possible interaction effects of opium consumption and other risk factors of cancers such as water pipe and cigarette consumption, and also study the effects of opium in never tobacco users.

From each case and control, a 10 mL sample of venous blood was collected for DNA extraction for future epidemiological studies to evaluate the association between opium use and genetic and molecular markers of cancers – a relationship which has not been studied before. Additionally, saliva samples were collected from head and neck cases and controls to study the prevalence of HPV and detect the most prevalent type of HPV among head and neck cancer cases in Iran. The data can also be combined with opium use, tobacco use, and other data for various epidemiologic studies.

The strengths of this case-control study are its large sample size, collection of detailed data on opium use and potential confounders, validation of the questionnaires, substantial training and supervision of the interviewers, high participation rates among cases and controls, careful selection of controls, and histologic confirmation of cancer diagnoses. Despite our best efforts, however, this study, as a case-control study, may still suffer from some biases like selection bias and some reporting bias. We did not choose a population-based control group because underreporting of opium use, as a sensitive matter, in the general population may be substantially higher than hospital visitors.[48]

In conclusions, we were able to successfully design and test various steps in conducting this large multi-center study. The full study with general questionnaires, food frequency questionnaires, and biological samples is by far the largest study of association on opium and cancer. The findings from this study are expected to make a significant contribution to advancing our knowledge on carcinogenicity of opium use. The study data can also be used to assess the effects of other potential risk factors of cancer, such as water-pipe smoking in Iran.

Authors' Contribution

All authors of this research paper have directly participated in the planning, execution, or analysis of the study. MH, HR, MM, MGH, EM, EE, AAH, AR, AR, ARM, PB, EW, FK, KZ, AM, FN, RS, AN, MS, AA, AN, RS, AS, MB, MM, EP, HP, SE, ON, SS, RS, AN contributed to the design and implementation of the study. KZ, FK, EP, as corresponding authors supervised the conduct and analyses of this study. All authors of this paper have read and approved the final version submitted. The contents of this manuscript have not been copyrighted or published previously. The contents of this manuscript are not now under consideration for publication elsewhere. The contents of this manuscript will not be copyrighted, submitted, or published elsewhere while acceptance by the journal is under consideration. There are no directly related manuscripts or abstracts, published or unpublished, by any authors of this paper.

Conflict of Interest Disclosures
None declared.

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Ethical Statement
The study was approved by the Ethics Committee of the National Institute of Medical Research Development (NIMAD) (Code: IR.NIMAD.REC.1394.027). All participants signed written inform consents, and data were handled confidentially.

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References
1. World drug report 2019 (United Nations publication, sales no. E.19.XI.8).
2. Amin-Esmaeili M, Rahimi-Movaghar A, Sharifi V, Hajebi A, Radgoodarzi R, Mojtabai R, et al. Epidemiology of illicit drug use disorders in Iran: prevalence, correlates, comorbidity and service utilization results from the Iranian Mental Health Survey. Addiction. 2016;111(10):1836-47. doi: 10.1111/add.13453.
3. Nalini M, Shakeri R, Poustchi H, Pourshams A, Etemadi A Islami F, et al. Long-term opiate use and risk of cardiovascular mortality: Results from the Golestan Cohort Study. Eur J Prev Cardiol. 2020; zwaa006. doi: 10.1093/eurjpc/zwaa006.
4. Alizadeh H, Naghibzadeh-Tahami A, Khanjani N, et al. Opium use and head and neck cancers: A matched case-control study in Iran. Asian Pac J Cancer Prev. 2020;21(3):783-790. doi: 10.31557/APJCP.2020.21.3.783.
5. Mousavi MRA, Damghani MA, Haghdoust AA, Khamesipour A. Opium and risk of laryngeal cancer. Laryngoscope. 2003;113(11):1939-43. doi: 10.1097/00005537-200311000-00016.
6. Mohebbi E, Hadji M, Rashidian H, Rezaianzadeh A, Marzban M, Haghdoost AA, et al. Opium use and the risk of head and neck squamous cell carcinoma. Int J Cancer. 2021;148(5):1066-1076. doi: 10.1002/ijc.33289.
7. Khoo R. Radiotherapy of carcinoma of the larynx. Ann Acad Med Singap. 1981;10(3):307-310.
8. Akbari M, Naghibzadeh TA, Khanjani N, et al. Opium as a risk factor for bladder cancer: A population-based case-control study in Iran. Arc Iran Med. 2015;18(9):567-571.
9. Afshari M, Janbabaei G, Bahrami MA, Moosazadeh M. Opium and bladder cancer: A systematic review and meta-analysis of the odds ratios for opium use and the risk of bladder cancer. PLoS One. 2017;12(6):e0178527. doi: 10.1371/journal.pone.0178527.
10. Sheikh M, Shakeri R, Poustchi H, Pourshams A, Etemadi A, Islami F, et al. Opium use and subsequent incidence of cancer: Results from the Golestan Cohort Study. Lancet Glob Health. 2020;8(5):e649-e660. doi: 10.1016/S2214-109X(20)30059-0.
11. MacLennan R, Da Costa J, Day NE, Law CH, Ng YK, Shanmugaratnam K. Risk factors for lung cancer in Singapore Chinese, a population with high female incidence rates. Int J Cancer. 1977;20(6):854-60. doi: 10.1002/ijc.2910200606.
12. Masjedi MR, Naghan PA, Taslimi S, Yousefifard M, Ebrahimi SM, Khosravi A, et al. Opium could be considered an independent risk factor for lung cancer: a case-control study. Respiration. 2013;85(2):112-8. doi: 10.1159/000338559.
13. Fahmy MS, Sadeghi A, Behmard S. Epidemiologic study of oral cancer in Fars province, Iran. Community Dent Oral Epidemiol. 1983;11(1):50-8. doi: 10.1111/j.1600-0528.1983.tb01354.x.
14. Bakhshaee M, Raziee HR, Afshari R, Amali A, Roopoosh M, Lotfizadeh A. Opium addiction and risk of laryngeal and esophageal carcinoma. Iran J Otorhinolaryngol. 2017;29(90):19-22.
15. Shakeri R, Malekzadeh R, Etemadi A, Nasrollahzadeh D, Aghcheli K, Sotoudeh M, et al. Opium: An emerging risk factor for gastric adenocarcinoma. Int J Cancer. 2013;133(2):455-61. doi: 10.1002/ijc.28018. Epub 2013 Feb 13.
16. Naghibzadeh TA, Khanjani N, Yazdi FV, Varzandeh M, Haghdoost AA. Opium as a risk factor for upper gastrointestinal cancers: A population-based case-control study in Iran. Arc Iran Med. 2014;17(1):2-6.
17. Malekzadeh MM, Khademi H, Pourshams A, Etemadi A, Poustchi H, Bagheri M, et al. Opium use and risk of mortality from digestive diseases: A prospective cohort study. Am J Gastroenterol. 2013;108(11):1757-65. doi: 10.1038/ajg.2013.336.
18. Ghadirian P, Stein GF, Gorodetzky C, Roberfroid MB, Mahon GA, Bartsch H, et al. Oesophageal cancer studies in the Caspian Littoral of Iran: Some residual results, including opium use as a risk factor. Int J Cancer. 1985;35(5):593-7. doi: 10.1002/ijc.2910350505.
19. Shakeri R, Kamangar F, Nasrollahzadeh D, Nouraie M, Khademi H, Etemadi A, et al. Is opium a real risk factor for esophageal cancer or just a methodological artifact? Hospital and neighborhood controls in case-control studies. PLoS One. 2012;7(3):e32711. doi: 10.1371/journal.pone.0032711.
20. Nasrollahzadeh D, Kamangar F, Aghcheli K, Sotoudeh M, Islami F, Abnet CC, et al. Opium, tobacco, and alcohol use in relation to oesophageal squamous cell carcinoma in a high-risk area of Iran. Br J Cancer. 2008;98(11):1857-63. doi: 10.1038/sj.bjc.6604369.
21. Moossavi S, Mohamadnejad M, Pourshams A, Poustchi H, Islami F, Sharafkhah M, et al. Opium use and risk of pancreatic cancer: A prospective cohort study. Cancer Epidemiol Biomarkers Prev. 2018;27(3):268-273. doi: 10.1158/1055-9965.EPI-17-0592.
22. Shakeri R, Kamangar F, Mohamadnejad M, Tabrizi R, Zamani F, Mohamadkhani A, et al. Opium use, cigarette smoking, and alcohol consumption in relation to pancreatic cancer. Medicine (Baltimore). 2016;95(28):e3922. doi: 10.1097/MD.0000000000003922.
23. Lankarani KB, Khosravizadegan Z, Naghibzadeh-Tahami A, Akbari M, Khodadost M, Honarvar B, et al. Opium use and risk of lower gastrointestinal cancers: Population-based case-control study in south of Iran. Int J Cancer Manag. 2017;10(6):e8227.
24. Naghibzadeh-Tahami A, Yazdi Feyzabadi V, Khanjani N, Ashrafi-Asgarabad A, Alizaeh H, Borhaninejad VR, et al. Can opium use contribute to a higher risk of colorectal cancers? A matched case-control study in Iran. Iran J Public Health. 2016;45(10):1322-1331.
25. IARC Monographs Vol 126 group. Carcinogenicity of opium consumption. Lancet Oncol. 2020;21(11):1407-1408. doi: 10.1016/S1470-2045(20)30611-2
26. Sheikh M, Kamangar F, Malekzadeh R. Fifty years of research

and one conclusion: Opium causes cancer. Arch Iran Med. 2020;23(11):757-760. doi: 10.34172/aim.2020.95.
27. Kamangar F, Shakeri R, Malekzadeh R, Islami F. Opium use: An emerging risk factor for cancer? Lancet Oncol. 2014;15(2):e69-77. doi: 10.1016/S1470-2045(13)70550-3
28. Estimation of the population size of substance and alcohol abusers in Iran. Research Center for Modeling in Health, Institute for Futures Studies in Health, Biostatistics and Epidemiology Department, Health School, Kerman University of Medical Sciences, Kerman, Iran; 2012:43-45.
29. Pourshams A, Khademi H, Malekshah AF, Islami F, Nouraei M, Sadjadi AR, et al. Cohort profile: The Golestan Cohort Study - a prospective study of oesophageal cancer in northern Iran. Int J Epidemiol. 2010;39(1):52-9. doi: 10.1093/ije/dyp161.
30. Vasheghani-Farahani A, Tahmasbi M, Asheri H, Ashraf H, Nedjat S, Kordi R. The Persian, last 7-day, long form of the international physical activity questionnaire: Translation and validation study. Asian J Sports Med. 2011;2(2):106-16. doi: 10.5812/asjsm.34781
31. Poustchi H, Eghtesad S, Kamangar F, Etemadi A, Keshtkar AA, Hekmatdoost A, et al. Prospective epidemiological research studies in Iran (the PERSIAN Cohort Study): Rationale, objectives, and design. Am J Epidemiol. 2018;187(4):647-655. doi: 10.1093/aje/kwx314.
32. Centers for Disease Control and Prevention. National health and nutrition examination survey: Anthropometry procedures manual. Atlanta, GA, USA: CDC; 2007.
33. Lee DH, Keum N, Hu FB, Orav EJ, Rimm EB, Sun Q, et al. Development and validation of anthropometric prediction equations for lean body mass, fat mass and percent fat in adults using the national health and nutrition examination survey (NHANES) 1999–2006. Br J Nutr. 2017;118(10):858-866. doi: 10.1017/S0007114517002665.
34. Aghaee-Afshar M, Khazaeli P, Behnam B, Rezazadehkermani M, Ashraf-Ganjooei N. Presence of lead in opium. Arch Iran Med. 2008;11(5):553-554.
35. Abnet CC, Saadatian-Elahi M, Pourshams A, Boffetta P, Feizzadeh A, Brennan P, et al. Reliability and validity of opiate use self-report in a population at high risk for esophageal cancer in Golestan, Iran. Cancer Epidemiol Biomarkers Prev. 2004;13(6):1068-70.
36. Rashidian H, Hadji M, Marzban M, Gholipour M, Rahimi-Movaghar A, Kamangar F, et al. Sensitivity of self-reported opioid use in case-control studies: Healthy individuals versus hospitalized patients. PLoS One. 2017;12(8):e0183017. doi: 10.1371/journal.pone.0183017.
37. Mohebbi E, Kamangar F, Rahimi-Movaghar A, Haghdoost AA, Etemadi A, Amirzadeh S, et al. An exploratory study of units of reporting opium use in Iran: Implications for epidemiologic studies. Arc Iran Med. 2019;22(10):541-545.
38. Nikfarjam A, Shokoohi M, Shahesmaeili A, Haghdoost AA, Baneshi MR, Haji-Maghsoudi S, et al. National population size estimation of illicit drug users through the network scale-up method in 2013 in Iran. Int J Drug Policy. 2016;31:147-52. doi: 10.1016/j.drugpo.2016.01.013.
39. Nakhaee N, Divsalar K, Meimandi MS, Dabiri S. Estimating the prevalence of opiates use by unlinked anonymous urine drug testing: A pilot study in Iran. Subst Use Misuse. 2008;43(3-4):513-20. doi: 10.1080/10826080701772348.
40. Ziaaddini H, Ziaaddini MR. The household survey of drug abuse in Kerman, Iran. J Applied Sci. 2005;5(2):380-382.
41. Bernaards CM, Twisk JW, Snel J, Van Mechelen W, Kemper HC. Is calculating pack-years retrospectively a valid method to estimate life-time tobacco smoking? A comparison between prospectively calculated pack-years and retrospectively calculated pack-years. Addiction. 2001;96(11):1653-61. doi: 10.1046/j.1360-0443.2001.9611165311.x.
42. Islami F, Kamangar F, Nasrollahzadeh D, Aghcheli K, Sotoudeh M, Abedi-Ardekani B, et al. Socio-economic status and oesophageal cancer: Results from a population-based case–control study in a high-risk area. Int J Epidemiol. 2009;38(4):978-88. doi: 10.1093/ije/dyp195
43. Caraceni A, Hanks G, Kaasa S, Bennett MI, Brunelli C, Cherny N, et al. Use of opioid analgesics in the treatment of cancer pain: Evidence-based recommendations from the EAPC. Lancet Oncol. 2012;13(2):e58-68. doi: 10.1016/S1470-2045(12)70040-2.
44. Zeppetella G, Davies AN. Opioids for the management of breakthrough pain in cancer patients. Cochrane Database Syst Rev. 2013;(10):CD004311. doi: 10.1002/14651858.CD004311.pub3.
45. Wang W. Illegal drug abuse and the community camp strategy in China. J Drug Educ. 1999;29(2):97-114. doi: 10.2190/J28R-FH8R-68A9-L288.
46. Shuster JJ, Cook B. Hospital or population controls: A discussion. J Chronic Dis. 1983;36(4):315-6. doi: 10.1016/0021-9681(83)90116-9.
47. Stavraky KM, Clarke EA. Hospital or population controls? An unanswered question. J Chronic Dis. 1983;36(4):301-7. doi: 10.1016/0021-9681(83)90113-3.
48. Mohebbi E, Rashidian H, Naghibzadeh Tahami A, Haghdoost AA, Rahimi-Movaghar A, Seyyedsalehi MS, et al. Opium use reporting error in case-control studies: neighborhood controls versus hospital visitor controls. Med J Islam Repub Iran. 2021;35(1):457-63.

PUBLICATION II

Opium use and the risk of head and neck squamous cell carcinoma.

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Opium use and the risk of head and neck squamous cell carcinoma

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Abbreviations: DMFT, decayed, missing and filled teeth index; HNSCC, head and neck squamous cell carcinoma; HPV, human papillomavirus; IROPICAN, Iran Opium and Cancer Study; ORs, odds ratios; 95% CI, 95% confidence intervals.

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Abstract

Scant evidence exists to support the association of opium use with head and neck cancer, limited to the larynx and oral cavity. In a multicenter case-control study—Iran Opium and Cancer study, we recruited 633 cases of head and neck squamous cell carcinoma (HNSCC) (254 lip and oral cavity, 54 pharynx, 327 larynx and 28 other subsites within the head and neck) and 3065 frequency-matched controls from April 2016 to April 2019. Odds ratios (ORs) for opium use and 95% confidence intervals (95% CIs) were obtained using mixed-effects logistic regression because of heterogeneity among centers. The adjusted OR (95% CI) for regular opium use was 3.76 (2.96-4.79) for all HNSCC combined. Strong dose-response effects were observed by frequency or amount of use, and duration of use. Regular opium uses significantly increased the risk of HNSCC of the pharynx, larynx and other subsites within the head and neck with OR (95% CI) of 2.90 (1.40-6.02), 6.55 (4.69-9.13) and 5.95 (2.41-14.71), respectively. The observed associations were significant even among never tobacco smokers (including cigarette and water-pipe smoking). Moreover, by the multiplicative interaction scale, the effect of opium use could be varied by cigarette smoking on HNSCC, 8.16 (6.20-10.74). For the first time, the current study showed opium users have an increased risk of several anatomic subsites of HNSCC.

KEYWORDS

drug-related disorders, morphine, neoplasm, opium, otorhinolaryngologic neoplasms

1 | INTRODUCTION

On a global scale, in 2018, an estimated 834 860 individuals developed new cancers of the lip, oral cavity, pharynx, and larynx, of whom 431 131 died due to these cancers.[1] A variety of etiological factors have been identified for head and neck cancers, including tobacco smoking, alcohol drinking, chewing betel quid, consumption of nitrosamine-rich foods and infection like human papillomavirus (HPV).[1]

Opium use is originated from the South Asian countries and the East Mediterranean including Iran. Although opium use is legally prohibited in Iran, it is the most commonly used drug.[2] Using opium has been identified as a risk factor for several cancers like cancers of

What's new?

Opium use has been associated with the risks of several cancers, but there is little data on whether opium contributes to head and neck cancer risk. Here, the authors conducted a multicenter case-control study, the Iran Opium and Cancer study (IROPICAN). They recruited 633 cases of head and neck squamous cell carcinoma and 3065 controls. The study drew from 10 provinces in Iran where opium use is most prevalent. They found that regular opium users have an elevated overall risk of HNSCC, and laryngeal cancer in particular.

bladder and lung.[3-5] However, there is very little data on the association of opium use and cancers of head and neck cancers.

A few case-control studies have found strong associations between the use of opium and the risk of laryngeal cancer.[6-10] The risk of supraglottic laryngeal cancer was also associated with a prescription of intravenous opioid in a case-control study.[11] Likewise, preliminary results from the Golestan cohort study also showed an increased risk of death due to laryngeal cancer in opium users.[12] Consistent with these findings, an ecological study in Iran showed a correlation between higher opium use and higher incidence rates of laryngeal cancer.[13] On the other hand, recent data from the national population-based cancer registry in Iran showed that the highest incidence rate of laryngeal cancer was reported from Kerman Province,[14] whereas the prevalence of opium consumption was higher than other regions.[15]

There were, however, serious limitations to previous studies of opium and laryngeal cancer. The earliest studies were conducted in the 1980s,[6,7] when the results were not typically adjusted for important confounding factors, such as tobacco smoking. Sample sizes were small—less than fifty cases—in some other studies. Furthermore, most previous studies were not primarily designed to study the effect of opium use on cancer, and as such, case and control selection and data collection methods were not optimal for this purpose.

We designed the Iran Opium and Cancer study (IROPICAN) to study the association of opium use and some types of cancers, among that opium use could be a possible and plausible risk factor, including cancers of the lip, oral cavity, pharynx and larynx. To overcome the limitations of the previous studies, we enrolled over 600 such cancer cases and 3000 controls in this study and optimized data collection and control selection methods during pilot studies.[16] The present report summarizes the findings of the IROPICAN study for all HNSCC cancers combined and cancers of the lip, oral cavity, pharynx and larynx, separately.

2 | METHODS

Data come from the IROPICAN study, a large multicenter case-control study conducted in 10 different provinces. These provinces were selected because the prevalence of opium use is relatively high in these regions.

2.1 | Case selection

Cases were incident head and neck squamous cell carcinomas (HNSCC) during April 2016 and April 2019, who referred to cancer care centers in the provinces. A team of trained researchers actively reviewed the admission and treatment notes of relevant wards (eg, surgical oncology wards) to identify potentially eligible HNSCC patients. The pathology reports were reviewed by the focal researchers and if needed they consulted clinicians to ensure the diagnosis. All head and neck cancer pathology reports that were not squamous cell types were excluded. HNSCC cases were further categorized by tumor sites according to the *International Classification of Diseases, Tenth Edition*.[17] We included cancers of lip (codes C00.0-C00.6, C00.8 and C00.9), oral cavity (codes C01.9, C02.0-C02.9, C03.0, C03.1, C03.9, C04.0, C04.1, C04.8, C04.9, C05.0- C05.2, C05.8, C05.9, C06.0-C06.2, C06.8, C06.8 and C06.9), salivary glands (codes C07.9, C08.0, C08.1, C08.08 and C08.9), tonsil (codes C09.0, C09.1, C09.8 and C09.9), oropharynx (codes C10.0-10.4, C10.8 and C10.9), nasopharynx (codes C11.0-C11.3, C11.8 and C11.09), hypopharynx (codes C13.0-C13.2, C13.8 and C13.9), other and ill-defined sites in lip, oral cavity and pharynx (codes C14.0, C14.2 and C14.8), nasal cavity and middle ear (codes C30.0 and C30.1), sinuses (codes C12.9, C31.0-C31.3, C31.8 and C31.9), larynx (codes C32.0-C32.3, C32.8 and C32.9), other and ill-defined sites (code C76.0) and head and neck cancers were overlapping or unspecified.[17] For analysis, the codes categorized to lip and oral cavity including codes C00-C08 and C14; pharynx codes C09-C11 and C13, larynx codes C32, and other subsites within head and neck codes C12, C31, C32 and C76. As only squamous cell carcinoma of head and neck was included, IROPICAN clinical consultant—head and neck surgeon—recommended combining the codes of other subsites (28 cases).

2.2 | Control selection

We selected at least four controls for each case: frequency matched by age, sex and place of residence. Potential controls were hospital visitors who were relatives or friends of hospitalized patients in either nononcology wards or who visited the hospital for any reason other than receiving treatment concurrently.[16] To reduce selection bias, emergency rooms and maternity wards were excluded for control recruitment because the referral pattern of the wards was more likely dependent on the residential area of residences, for example, accident injured persons referred to the closest emergency room (EMR), furthermore, drug and alcohol users increase car collision rate.[18,19] The controls were recruited in the same hospitals as the cases or in comparable referral hospitals of the catchment area. To be eligible, the controls had to be also free of any history of cancer reported by themselves.

We chose controls from among hospital visitors every day by a predefined protocol, previously we found that the prevalence of self-reported regular opioid use among hospital visitors was comparable to the general population.[16] Moreover, a high level of sensitivity (77%) for self-reporting of opium use among hospital visitor controls supported the use of hospital visitors as controls.[16]

2.3 | Data collection

A team of trained interviewers administered a structured questionnaire to both cases and controls. This questionnaire included detailed data on demographics, history of opium and tobacco use, history of alcohol drinking, oral health and socioeconomic status. The same team of trained interviewers also conducted physical exams including standardized measurements of height, weight and blood pressure

(both cases and controls). In each center, an assigned nurse collected blood and saliva samples using a predefined protocol.

2.4 | Opium exposure measurement

We collected a detailed history of opium use including the age of starting and ending use, frequency of use, the typical amount of use, types of opiates and routes of administration. In Iran, opium is used in various forms, including Teriak (crude opium), Sukhteh (remnants of smoked opium or dross) and Shireh (opium juice, an opium product usually made by boiling Teriak or Sukhteh with water, filtering the mixture several times and then evaporating the filtrate).[20] Because very few numbers of Sukhteh users (four users), we merged Sukhteh and Teriak. Regular opium user was defined as using opium at least once a week for at least 6 consecutive months. Route of opium use was also inquired since all forms of opium can be used via smoking and oral ingestion.

A measure of cumulative opium use in a lifetime was defined as the sum of the amount of opium use (gram per day) multiplied by the amount of use in each duration of opium use in a lifetime (gram-year). Another approach to explore dose-response was the multiplication of frequency of use and duration of that period of opium use (frequency-year).[21] The other metric of opium is the average intensity of opium use. The average intensity of opium was calculated by dividing the cumulative opium use to the duration of that period. All of the measures were categorized into three groups by the tertile of the control group.

Reverse causality is an important concern for the association of opium use and any cancer, since the patients may start using opium to alleviate their pain, which could be a prodromal symptom of the subsequently diagnosed cancer. Thus, we disregarded opium use in those who started 3 years before diagnosis. Consequently, six patients were categorized as the nonopium user to reduce reverse causation in all analyses and tables.

In the validation study, we found that current opium use had limited sensitivity when tested against morphine in urine.[16] Hence, it is plausible that the regular use of opium subject to more nondifferential misclassification (information bias). To overcome information bias—underreporting of opium use—we calculated odds ratios (ORs) for a range of 0.5 to 0.9 of self-reporting in cases and controls and drawing a surface plot. Besides, based on point estimation of the sensitivity of reporting opium use, for cases, we considered 0.77 and 0.68 for controls and corrected adjusted ORs are reported.

2.5 | Statistical analysis

All statistical analyses were conducted using Stata, version 14 (Stata Corp, College Station, Texas). Frequencies and percentages were calculated for categorical variables. As we recruited the subjects from 10 different centers, heterogeneity between centers was tested (*P* heterogeneity) and mixed-effects logistic regression models with random intercept by the center of the study was applied to estimate the association of opium use with HNSCC status (OR with 95% confidence interval). We present adjusted ORs, with the latter adjusted for potential confounders including age; gender; place of residence (center/noncenter); cigarette smoking (pack-year); water-pipe smoking (head-year); alcohol drinking (regular drinkers/nonregular drinkers); decayed, missing and filled teeth (DMFT) index as an indicator of oral health; and socioeconomic status. Socioeconomic status was determined using principal components analysis, by combining years of education (continuous variable) and ownership of some assets (dichotomous variables; washing machine, freezer, personal computer, sofa, vacuum cleaner, dishwasher, split air conditioner, owned house, owned car). Analyses were conducted for all HNSCC, as well as for four main anatomic subsites as introduced earlier.

Since tobacco and alcohol are two major HNSCC risk factors, we also conducted analyses restricted to those who are never tobacco smokers (Supplementary Table 2).

The *P* value for multiplicative interaction was obtained employing a Wald test of the interaction coefficient in the logistic regression.

3 | RESULTS

A total of 663 HNSCC cases (254 lip and oral cavity, 54 pharynx, 327 larynx and 28 other subsites within head and neck) and 3065 frequency-matched controls were enrolled in the IROPICAN study. Table 1 shows the distribution of demographic and habit variables in cases and controls. Among cases, approximately 75% were men, 73% were capital city residents and the median age at recruitment was 58 (25th centile 50 and 75th centile 66 years). The corresponding numbers in controls were 68%, 78% and 57 (49-64) years, respectively. Cases were more likely than controls to smoke, consume alcohol, have lower SES and have poorer oral health (Table 1). Two percent of cases and 14% of controls nonresponses were refusals, mostly because of donating blood. No difference in age and gender was observed between participants and nonrespondents.

Regular opium use was strongly associated with a higher risk of HNSCC. Table 2 shows the results for the association of regular opium use with all HNSCC combined. The adjusted OR (95% CI) for regular opium use was 3.76 (2.96-4.79). There was a strong dose-response association when associations were investigated by the duration of use, cumulative use and frequency of opium. For example, the OR (95% CI) was 2.06 (1.22-3.47) for those who had an above the third tertile of cumulative opium use (≥24.5 gram-year) among users, as compared to an OR of 2.27 (1.36-3.78) for those with the second tertile of cumulative use (3.7-24.5 gram-year).

Both common types of opium used, that is, crude opium (Teriak) and opium juice (Shireh) were strongly associated with higher HNSCC risk. However, Shireh with an OR (95% CI) of 7.17 (4.44-11.58) had a stronger association with HNSCC than Teriak [3.40 (2.64-4.37)]. Both routes of opium use, that is, oral ingestion and smoking were strongly associated with a higher risk of HNSCC. However, oral ingestion, with an OR (95% CI) of 8.33 (4.67-14.58) was more strongly associated

TABLE 1 Distribution of demographic and habits for head and neck squamous cell carcinoma cases and controls

	HNSCC cases[a] N (%)	Controls N (%)	*P* value
Total	663	3065	
Age			.36
≤29	9 (1.36)	25 (0.82)	
30 to 39	45 (6.79)	246 (8.03)	
40 to 49	100 (15.08)	517 (16.87)	
50 to 59	213 (32.13)	982 (32.04)	
60 to 69	203 (30.62)	923 (30.11)	
≥70	93 (14.03)	372 (12.14)	
Gender			<.0001
Male	499 (75.26)	2071 (67.57)	
Female	164 (24.74)	994 (32.43)	
Place of residence			.007
Capital city	487 (73.45)	2399 (78.27)	
Noncapital city	176 (26.55)	666 (21.73)	
Opium use[b]			<.0001
Nonregular user[c]	368 (55.51)	2664 (86.92)	
Regular user[d]	295 (44.49)	401 (13.08)	
Cigarette smoking			<.0001
Nonregular user	292 (44.04)	2220 (72.43)	
Regular user[e]	371 (55.96)	845 (27.57)	
Pack-years of cigarette smoking	20.82 ± 29.84	5.33 ± 13.14	<.0001
Water-pipe smoking			
Nonregular-user	602 (90.80)	2858 (93.25)	.02
Regular user[f]	61 (9.20)	207 (6.75)	
Head-years of water-pipe smoking	48.61 ± 69.12	35.65 ± 65.56	.08
Alcohol drinking			<.0001
Nonuser	617 (93.06)	2947 (96.15)	
Regular user[g]	46 (6.94)	118 (3.85)	
Socioeconomic status[h]			<.0001
Low	400 (60.33)	1440 (46.98)	
High	263 (39.67)	1625 (53.02)	
DMFT index[i]			<.0001
Poor	489 (73.76)	1510 (49.27)	
Good	174 (26.24)	1555 (50.73)	

[a]HNSCC: head and neck squamous cell carcinoma. Cases and controls were frequency matched on age, gender and place of residence.
[b]Regular opium use: using opium at least once a week for at least a 6-month consecutive period during the lifetime.
[c]Nonuser included nonregular users.
[d]After reclassifying opium users who started opium use within 3-year prior cancer diagnosis.
[e]Regular cigarette smoking: smoking a cigarette per week for at least a 6-month consecutive period during the lifetime.
[f]Regular water-pipe smoking: smoking a head of water pipe per week for at least a 6-month consecutive period during the lifetime.
[g]Regular alcohol drinking: drinking any types of alcohol at least once a week for at least a 6-month consecutive period during the lifetime.
[h]We used the median in control subjects as the dividing cut point.
[i]DMFT index: decayed, missing and filled teeth index.

TABLE 2 The associations of opium use with head and neck squamous cell carcinoma

	HNSCC cases[a] N (%)	Controls N (%)	Adjusted OR[b] (95% CI[c])
Regular opium use[d]			
Nonuser[e]	368 (55.51)	2664 (86.92)	Referent
Regular user[f]	295 (44.49)	401 (13.08)	3.76 (2.96-4.79)
P for heterogeneity			<.0001
Duration of opium use (year)			
First tertile (≤11)	51 (17.29)	143 (35.66)	Referent
Second tertile (12–23)	101 (34.24)	127 (31.67)	1.68 (1.04-2.72)
Third tertile (≥24)	143 (48.47)	131 (32.67)	2.52 (1.55-4.11)
P trend[g]			<.0001
P for heterogeneity			<.0001
Cumulative use[h] (gram-year)			
First tertile (≤3.6)	38 (12.88)	134 (33.42)	Referent
Second tertile (3.7–24.5)	104 (35.25)	134 (33.42)	2.27 (1.36-3.78)
Third tertile (≥24.5)	153 (51.86)	133 (33.17)	2.06 (1.22-3.47)
P trend			.022
P for heterogeneity			<.0001
Frequency-year[i]			
First tertile (≤8)	30 (10.17)	138 (34.41)	Referent
Second tertile (8.1-22)	52 (17.63)	130 (32.42)	1.70 (0.97-2.99)
Third tertile (≥23)	213 (72.20)	133 (33.17)	5.09 (3.05-8.47)
P trend			<.0001
P for heterogeneity			<.0001
Average intensity (gram/day)			
First tertile (≤0.4)	62 (21.02)	150 (37.41)	Referent
Second tertile (0.5-2)	110 (37.29)	118 (29.43)	1.33 (0.83-2.13)
Third tertile (≥2)	123 (41.69)	133 (33.17)	0.88 (0.53-1.44)
P trend			.46
P for heterogeneity			<.0001
Type of opium used			
Nonuser	368 (55.51)	2664 (86.92)	Referent
Crude opium (Teriak)	238 (35.90)	360 (11.75)	3.40 (2.64-4.37)
Opium juice (Shireh)	57 (8.60)	41 (1.34)	7.17 (4.44-11.58)
P for heterogeneity			<.0001
Route of opium use			
Nonuser	368 (55.51)	2664 (86.92)	Referent
Only smoking	168 (25.34)	337 (11.00)	2.66 (2.03-3.47)
Only oral ingestion	35 (5.28)	28 (0.91)	8.33 (4.67-14.85)
Both routes	92 (13.88)	36 (1.17)	12.96 (8.14-20.62)
P for heterogeneity			<.0001

[a]HNSCC: head and neck squamous cell carcinoma. Cases and controls were frequency matched on age, gender and place of residence.
[b]Random-effect odds ratio. Adjusted for age (categorical), gender (categorical), place of residence (categorical), pack-years of cigarette smoking (continuous), head-years of water-pipe smoking (continuous), regular alcohol drinking (categorical), socioeconomic status (categorical) and oral health (DMF index: continuous). Likelihood heterogeneity test by the center.
[c]95% CI: 95% confidence interval.
[d]Regular opium use: using opium at least once a week for at least a 6-month consecutive period during the lifetime.
[e]Nonuser included nonregular users.
[f]After reclassifying opium users who started opium use within 3-year prior cancer diagnosis.
[g]*P* trend: *P* values for trend were obtained from adjusted models by assigning values of 1, 2 and 3 to low use (T1), moderate use (T2) and high use (T3), respectively.
[h]Cumulative use: total frequency of opium use (per day) multiplied amount (gram) of opium and total duration (year).
[i]Frequency-year: total frequency of opium use (per day) multiplied total duration (year).

TABLE 3 The associations of opium use with head and neck squamous cell carcinoma by anatomical subsites

		Lip and oral cavity (254 cases)		Pharynx (54 cases)		Larynx (327 cases)		Other subsites[a] (28 cases)	
	Controls N (%)	Cases N (%)	Adjusted OR[b] (95% CI)	Cases N (%)	Adjusted OR (95% CI)	Cases N (%)	Adjusted OR (95% CI)	Cases N (%)	Adjusted OR (95% CI)
Regular opium use[c]									
Nonuser[d]	2664 (86.92)	221 (87.01)	Referent	37 (68.52)	Referent	96 (29.36)	Referent	14 (50.00)	Referent
Regular user[e]	401 (13.08)	33 (12.99)	1.53 (0.97-2.41)	17 (31.48)	2.90 (1.40-6.02)	231 (70.64)	6.55 (4.69-9.13)	14 (50.00)	5.95 (2.41-14.71)
P for heterogeneity			.28		<.0001		<.0001		<.0001
Duration of opium use (year)									
First tertile (≤11)	143 (35.66)	8 (24.24)	Referent	5 (29.41)	Referent	35 (15.15)	Referent	3 (21.43)	Referent
Second tertile (12–23)	127 (31.67)	11 (33.33)	1.01 (0.37-2.76)	5 (29.41)	0.93 (0.23-3.75)	80 (34.63)	1.91 (1.10-3.31)	5 (35.71)	1.89 (0.35-10.05)
Third tertile (≥24)	131 (32.67)	14 (42.42)	2.09 (0.75-5.80)	7 (41.8)	1.9 (0.4-8.6)	116 (50.22)	2.71 (1.56-4.68)	6 (42.86)	2.96 (0.55-15.91)
P trend[f]			.15		.40		<.0001		.20
P for heterogeneity			.43		<.0001		<.0001		
Cumulative use[g] (gram-year)									
First tertile (≤3.6)	134 (33.42)	7 (21.22)	Referent	4 (23.53)	Referent	26 (11.26)	Referent	1 (7.14)	Referent
Second tertile (3.7-24.4)	134 (33.42)	13 (39.39)	1.52 (0.56-4.13)	6 (35.29)	1.35 (0.31-5.83)	77 (33.33)	2.32 (1.28-4.20)	8 (57.14)	9.79 (1.06-89.78)
Third tertile (≥24.5)	133 (33.16)	13 (39.39)	1.24 (0.44-3.43)	7 (41.18)	1.07 (0.22-5.08)	128 (55.41)	2.29 (1.26-4.16)	5 (35.72)	6.71 (0.65-68.99)
P trend			.73		.95		.01		.13
P for heterogeneity			.46		<.0001		<.0001		<.0001
Frequency-year[h]									
First tertile (≤8)	138 (34.41)	11 (33.33)	Referent	3 (17.65)	Referent	14 (6.06)	Referent	2 (14.29)	Referent
Second tertile (8.1–22)	130 (32.42)	5 (15.15)	0.41 (0.13-1.27)	3 (17.65)	0.99 (0.17-5.54)	43 (18.61)	3.38 (1.63-6.99)	1 (7.14)	0.31 (0.02-4.06)
Third tertile (≥23)	133 (33.17)	17 (51.52)	1.24 (0.52-2.95)	11 (64.70)	3.24 (0.76-13.71)	174 (75.32)	9.05 (4.62-17.71)	11 (78.57)	5.53 (1.03-29.66)
P trend			.53		.07		<.0001		.02
P for heterogeneity			.19		<.0001		<.0001		<.0001

	Controls N (%)	Lip and oral cavity (254 cases)		Pharynx (54 cases)		Larynx (327 cases)		Other subsites [a] (28 cases)	
		Cases N (%)	Adjusted OR[b] (95%CI)	Cases N (%)	Adjusted OR (95% CI)	Cases N (%)	Adjusted OR (95% CI)	Cases N (%)	Adjusted OR (95% CI)
Average intensity (gram/day)									
First tertile (≤0.4)	150 (37.40)	7 (21.21)	Referent	5 (29.41)	Referent	44 (19.05)	Referent	6 (42.86)	Referent
Second tertile (0.5–2)	118 (29.43)	15 (45.45)	2.28 (0.869-6.03)	8 (47.06)	1.63 (0.48-6.51)	83 (35.93)	1.27 (0.74-2.16)	4 (28.57)	0.80 (0.19-3.34)
Third tertile (≥2)	133 (33.17)	11 (33.34)	1.12 (0.39-3.19)	4 (23.53)	0.41 (0.07-2.26)	104 (45.02)	0.92 (0.53-1.60)	4 (28.57)	0.82 (0.19-3.42)
P trend			.96		.26		.62		.77
P for heterogeneity			.52		<.0001		<.0001		<.0001

TABLE 3 (Continued)

		Lip and oral cavity (254 cases)		Pharynx (54 cases)		Larynx (327 cases)		Other subsites [a] (28 cases)	
	Controls N (%)	Cases N (%)	Adjusted OR[b] (95%CI)	Cases N (%)	Adjusted OR (95% CI)	Cases N (%)	Adjusted OR (95% CI)	Cases N (%)	Adjusted OR (95% CI)
Type of opium used									
Nonuser	2664 (86.92)	221 (87.01)	Referent	37 (68.52)	Referent	96 (29.36)	Referent	14 (50.00)	Referent
Crude opium (Teriak)	360 (11.75)	28 (11.02)	1.41 (0.87-2.27)	15 (27.78)	2.81 (1.32-5.97)	182 (55.66)	5.77 (4.09-8.15)	13 (46.43)	6.04 (2.43-15.05)
Opium juice (Shireh)	41 (1.33)	5 (1.97)	2.90 (1.05-7.97)	2 (3.70)	3.77 (0.80-17.68)	49 (14.98)	12.69 (7.25-22.22)	1 (3.57)	4.83 (0.55-41.97)
P for heterogeneity			.37		<.0001		<.0001		.002
Route of opium use							<0.0001		<0.0001
Nonuser	2664 (86.92)	221 (87.01)	Referent	37 (68.52)	Referent	96 (29.36)	Referent	14 (50.00)	Referent
Only smoking	337 (11.00)	20 (7.87)	1.09 (0.64-1.86)	15 (27.78)	3.04 (1.43-6.47)	125 (38.23)	4.28 (2.98-6.14)	8 (28.58)	3.97 (1.44-10.99)
Only oral ingestion	28 (0.91)	6 (2.36)	4.25 (1.45-11.69)	1 (1.85)	2.67 (0.33-21.57)	25 (7.65)	17.17 (8.44-34.91)	3 (10.71)	17.92 (4.32-74.26)
Both routes	36 (1.17)	7 (2.76)	5.10 (2.41-12.89)	1 (1.85)	1.74 (0.21-14.26)	81 (24.77)	25.11 (14.55-43.33)	3 (10.71)	11.96 (2.83-50.52)
P for heterogeneity			.17		<.0001		<.0001		<.0001

[a]Other subsites included pyriform sinus, nasal cavity, and middle ear, accessory sinuses, and head and neck, NOS.
[b]Random-effect odds ratio. Adjusted for age (categorical), gender (categorical), place of residence (categorical), pack-years of cigarette smoking (continuous), head-years of water-pipe smoking (continuous), regular alcohol drinking (categorical), socioeconomic status (categorical) and oral health (DMF index: continuous). Likelihood heterogeneity test by the center.
[c]Regular opium use: using opium at least once a week for at least a 6-month consecutive period during the lifetime.
[d]Nonuser included nonregular users.
[e]After reclassifying opium users who started opium use within 3-year prior cancer diagnosis.
[f]*P* trend: *P* values for trend were obtained from adjusted models by assigning values of 1, 2 and 3 to low use (T1), moderate use (T2) and high use (T3), respectively.
[g]Cumulative use: total frequency of opium use (per day) multiplied amount (gram) of opium, then multiplied total duration (year).
[h]Frequency-year: total frequency of opium use (per day) multiplied total duration (year).

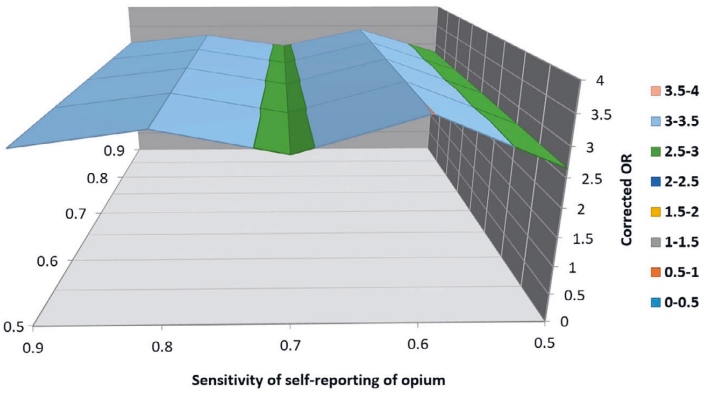

FIGURE 1 The surface plot of corrected OR and sensitivity of self-reporting of opium ranged from 0.50 to 0.90 in cases and controls

with HNSCC than smoking [2.66 (2.03-3.47)]. The strongest associations were seen in those who used both routes [12.96 (8.14-20.62)].

Regular opium use significantly increased the risk of HNSCC of the pharynx, larynx and other subsites within HNSCC, with OR (95% CI) of 2.90 (1.4-6.02), 6.55 (4.69-9.13) and 5.95 (2.41-14.71), respectively (Table 3). A dose-response association was seen for various metrics of opium use with the risk of the larynx and other subsites within HNSCC. The association with regular opium use varied between anatomical subsites of the larynx; the OR (95%CI) for supraglottis was 18.27 (8.23-40.53), glottis 6.20 (3.61-10.63), larynx, NOS 4.38 (2.49-7.70) and other subsites of larynx 7.89 (4.21-14.77) (Supplementary Table 1).

By contrast, no statistically significant association was observed for lip and oral cavity squamous cell carcinoma, with an OR (95% CI) of 1.53 (0.97-2.41) for regular opium use and of 1.24 (0.44-3.43) of cumulative use.

The association of opium use with HNSCC risk persisted among never tobacco smokers (Supplementary Table 2). The OR (95% CI) for the association between opium use and HNSCC in never smokers was 5.17 (3.26-8.21).

Tests for interaction were significant on the multiplicative scale for HNSCC combined that was 8.16 (6.20-10.74), lip and oral cavity 1.97 (1.21-3.19), pharynx 4.88 (2.09-11.41), larynx 28.78 (17.92, 46.21) and other anatomic subsites 5.53 (1.70-18). Hence, the effect of opium could be varied by cigarette smoking.

According to our sensitivity study, the impact of the sensitivity of self-reporting of opium, 0.77 in cases and 0.68 in controls indicated that the association of HNSCC combined and regular opium use was still significant, corrected OR was 2.48 (2.05 to 2.98). In addition, the surface plot of corrected OR and sensitivity of self-reporting of opium ranged from 0.50 to 0.90 in both groups showed that the null zone of crude OR did not cross (Figure 1).

4 | DISCUSSION

With over 600 cases and 3000 controls, this study is by far the largest study of opium use and HNSCC conducted to date. Opium use is associated with a remarkably increased risk of HNSCC and some anatomic HNSCC subsites, including cancers of the pharynx, larynx and the other subsite groups. The risk of cancers of the lip and oral cavity was not increased in regular opium users.

Our findings suggest a causal relationship. We adjusted for important potential confounders, including age, gender, cigarette smoking, water-pipe smoking, alcohol consumption and socioeconomic status (SES); nevertheless, the results remained statistically significant. Similarly, when we restricted the analyses to certain subgroups, such as never cigarette and water-pipe smokers, and the associations remained significant and strong. There was a clear dose-response association with duration of use, frequency of use and cumulative use. Compared to more frequent opium users, those who used opium more than 22 times a year were five times more likely to have HNSCC than those used opium eight times a year.

Our findings of an increased risk of HNSCC are in agreement with those of five prior case-control studies.[6,8-11] The only study that differed was a cross-sectional study of 44 laryngeal cancers. It showed no association of opium dependency and the pattern of laryngeal anatomic regions.[22] The overall strong consistency of association is again in favor of a causal relationship. The increased risk of HNSCC associated with opium use varied by subsite. The association was particularly strong for laryngeal cancer, which is consistent with previous literature studies.[6,8-10] The potential mechanisms for variations across anatomic subsites are unclear.

To rule out the effect of reverse causality on the association of opium use and risk of HNSCC, we reclassified opium users who started opium use 3 years before diagnosis as nonusers. We chose 3 years, which is longer than most other case-control studies considered, to be sure that even early manifestations of the HNSCC (such as a recurrent wound, coughs) were not alleviated using opium, particularly for slow-growing tumors.[23] There is other evidence against reverse causality. The large majority of opium users, in both cases and controls, had been using opium for quite a long time; the median duration of regular opium use was approximately 20 years.

Of note, both Teriak and Shireh, the two major types of opium used by our study participants, were associated with a higher risk of

HNSCC. Likewise, both oral ingestion and smoking of opium were associated with higher HNSCC risk. Both opium ingestion and opium smoking have been associated with higher risk of several cancers, including cancers of the bladder and other sites that do not come into direct contact with opium products.[3,24] The significance of these findings is not entirely clear, but it may show that it is really the alkaloids in the opium that are the major drivers of carcinogenicity.

Our study has several strengths, including its large sample size; histologic confirmation of all cases; investigating the association by anatomic subsites; choosing hospital visitor controls, which were shown to be the appropriate control group to study the effects of opium[16]; using trained investigators and validated structured questionnaires. The strict control of confounders, by limiting the analyses to never tobacco smokers, is another advantage of this study. Using hospital visitor controls turned out to be the favored option among potential control groups tested in validation study.[16] Especially, they showed the most accurate reporting of opium use and also a high response rate.

Our study may have some limitations too. Like other case-control studies, information bias may be a source of biased results. To alleviate this problem, we designed an extensive questionnaire and devised the order of the questions such that neither the interviewers nor the study participants had any preconceived notion that opium was the main study exposure. Likewise, during the training, we did not emphasize the importance of opium in this study. We also tried to minimize interviewer bias using a comprehensive protocol of interviewer training, data collection and monthly reviewing the protocols.

Despite the overall large sample size, some anatomic subsites had small sample sizes. We did not have data on HPV infections, an important risk factor for oropharyngeal SCC, which may also be associated with drug use.[25-28] However, only about 7% of Iranian women are positive for cervical HPV[29,30] and the rates are likely much lower for oropharyngeal HPV. Furthermore, the association of opium and HNSCC were strong among men, who constituted more than two-thirds of our study population and in whom the prevalence of HPV is far lower than women.[31] Therefore, it is unlikely that HPV is a major confounder.

5 | CONCLUSION

In conclusion, we found evidence of a positive association between opium use and the risk of HNSCC, overall and by most anatomical subsites. These findings as add to studies finding cancers of other organs such as bladder, esophagus and lung related to opium use, suggesting that opium use is an important carcinogen.

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CONFLICT OF INTEREST

All authors disclosed no financial and personal relationships with other people and organizations that could inappropriately influence (bias) their work.

DATA AVAILABILITY STATEMENT

The dataset used in this study is held securely in the coded format at Cancer Research Center (CRC) of Iran. Although data sharing agreements prohibit CRC from making the dataset publicly available, access may be granted to those who meet the conditions for confidential access and on reasonable request via the corresponding author's email (kzendeh@sina.tums.ac.ir).

ETHICS STATEMENT

The study was approved by the Institutional Review Boards of the National Institute for Medical Research Development (IR.NIMAD.REC.1394.027). Written informed consent was obtained.

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REFERENCES

1. Shield KD, Ferlay J, Jemal A, et al. The global incidence of lip, oral cavity, and pharyngeal cancers by subsite in 2012. *CA Cancer J Clin*. 2017;67:51-64.
2. Merz F. United nations office on drugs and crime: world drug report 2017. 2017. *SIRIUS-Zeitschrift für Strategische Analysen*. 2018;2:85-86.
3. Kamangar F, Shakeri R, Malekzadeh R, Islami F. Opium use: an emerging risk factor for cancer? *Lancet Oncol*. 2014;15:e69-e77.
4. Masjedi MR, Naghan PA, Taslimi S, et al. Opium could be considered an independent risk factor for lung cancer: a case-control study. *Respiration*. 2013;85:112-118.
5. Afshari M, Janbabaei G, Bahrami MA, Moosazadeh M. Opium and bladder cancer: a systematic review and meta-analysis of the odds ratios for opium use and the risk of bladder cancer. *PLoS One*. 2017;12:e0178527.
6. Khoo R. Radiotherapy of carcinoma of the larynx. *Ann Acad Med Singapore*. 1981;10:307-310.
7. Fahmy MS, Sadeghi A, Behmard S. Epidemiologic study of oral cancer in Fars Province, Iran. *Community Dent Oral Epidemiol*. 1983;11:50-58.
8. Mousavi MR, Damghani MA, Haghdoust AA, Khamesipour A. Opium and risk of laryngeal cancer. *Laryngoscope*. 2003;113:1939-1943.
9. Bakhshaee M, Raziee HR, Afshari R, Amali A, Roopoosh M, Opium Addiction LA. Risk of laryngeal and esophageal carcinoma. *Iranian J Otorhinolaryngol*. 2017;29:19-22.
10. Alizadeh H, Naghibzadeh Tahami A, Khanjani N, et al. Opium use and head and neck cancers: a matched case-control study in Iran. *Asian Pac J Cancer Prev*. 2020;21:783-790.
11. Shoffel-Havakuk H, Cohen O, Slavin M, Haimovich Y, Halperin D, Lahav Y. Intravenous opioid drug abuse as an independent risk factor for supraglottic squamous cell carcinoma-a case-control study. *Clinical Otolaryngol*. 2018;43:456-462.

12. Rahmati A, Shakeri R, Khademi H, et al. Mortality from respiratory diseases associated with opium use: a population-based cohort study. *Thorax*. 2017;72:1028-1034.
13. Rashidian H, Zendehdel K, Kamangar F, Malekzadeh R, Haghdoost AA. An ecological study of the association between opiate use and incidence of cancers. *Addict Health*. 2016;8:252-260.
14. Roshandel G, Ghanbari-Motlagh A, Partovipour E, et al. Cancer incidence in Iran in 2014: results of the Iranian National Population-based Cancer Registry. *Cancer Epidemiol*. 2019;61:50-58.
15. Nikfarjam A, Shokoohi M, Shahesmaeili A, et al. National population size estimation of illicit drug users through the network scale-up method in 2013 in Iran. *Int J Drug Policy*. 2016;31:147-152.
16. Rashidian H, Hadji M, Marzban M, et al. Sensitivity of self-reported opioid use in case-control studies: healthy individuals versus hospitalized patients. *PLoS One*. 2017;12:e0183017.
17. Fritz AG. *International classification of diseases for oncology: ICD-O*. Lyon, France: World Health Organization; 2013.
18. Asbridge M, Hayden JA, Cartwright JL. Acute cannabis consumption and motor vehicle collision risk: systematic review of observational studies and meta-analysis. *BMJ*. 2012;344:e536.
19. Wickens CM, Mann RE, Ialomiteanu AR, et al. The impact of medical and non-medical prescription opioid use on motor vehicle collision risk. *Transp Res Part F Traffic Psychol Behav*. 2017;47:155-162.
20. Amin-Esmaeili M, Rahimi-Movaghar A, Sharifi V, et al. Epidemiology of illicit drug use disorders in Iran: prevalence, correlates, comorbidity and service utilization results from the Iranian mental health survey. *Addiction*. 2016;111:1836-1847.
21. Mohebbi E, Kamangar F, Rahimi-Movaghar A, et al. An exploratory study of units of reporting Opium Usein Iran: implications for epidemiologic studies. *Arch Iran Med*. 2019;22:541-545.
22. Dabirmoghaddam P, Taheri AK, Ghazavi H, Ebrahimnejad S, Karimian Z. Does opium dependency affect the pattern of involvement in laryngeal cancer? *Iranian J Otorhinolaryngol*. 2016;28:425.
23. Timar J, Csuka O, Remenar E, Repassy G, Kasler M. Progression of head and neck squamous cell cancer. *Cancer Metastasis Rev*. 2005;24:107-127.
24. Sheikh M, Shakeri R, Poustchi H, et al. Opium use and subsequent incidence of cancer: results from the Golestan cohort study. *Lancet Glob Health*. 2020;8:e649-e660.
25. Parks KA, Collins RL, Derrick JL. The influence of marijuana and alcohol use on condom use behavior: findings from a sample of young adult female bar drinkers. *Psychol Addict Behav*. 2012;26:888-894.
26. Bryan AD, Schmiege SJ, Magnan RE. Marijuana use and risky sexual behavior among high-risk adolescents: trajectories, risk factors, and event-level relationships. *Dev Psychol*. 2012;48:1429-1442.
27. Tetrault JM, Fiellin DA, Niccolai LM, Sullivan LE. Substance use in patients with sexually transmitted infections: results from a national US survey. *Am J Addict*. 2010;19:504-509.
28. Jones AA, Striley CW, Cottler LB. Prescription opioid use, illicit drug use, and sexually transmitted infections among participants from a community engagement program in north Central Florida. *J Subst Abus*. 2017;22:90-95.
29. Khodakarami N, Clifford GM, Yavari P, et al. Human papillomavirus infection in women with and without cervical cancer in Tehran. *Iran Intern J Cancer*. 2012;131:E156-E161.
30. Khorasanizadeh F, Hassanloo J, Khaksar N, et al. Epidemiology of cervical cancer and human papilloma virus infection among Iranian women—analyses of national data and systematic review of the literature. *Gynecol Oncol*. 2013;128:277-281.
31. Seifi S, Kermani IA, Dolatkhah R, et al. Prevalence of oral human papilloma virus in healthy individuals in East Azerbaijan province of Iran. *Iran J Public Health*. 2013;42:79.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

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PUBLICATION III

Opium Use and Risk of Bladder Cancer: A Multi-center Case-Referent Study in Iran

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Original article

Cancer

Opium use and risk of bladder cancer: a multi-centre case-referent study in Iran

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Abstract

Background: Bladder cancer (BC) is the 10th most common type of cancer worldwide and the fourth most common type of cancer in Iran. Opium use is considered as one of the risk factors for BC. We aim to assess the association between various parameters of opium use, which in Iran is mainly ingested or smoked in various forms, and the risk of BC.

Method: In this multi-centre case-referent study in Iran, 717 BC cases and 3477 referents were recruited to the study from May 2017 until July 2020. Detailed histories of opium use (duration, amount, frequency) and potential confounders were collected by trained interviewers. Multivariable unconditional logistic regression models were used to measure adjusted odds ratio (OR) and 95% confidence intervals (CI). The ORs were adjusted for age, gender, place of residence and pack-years of cigarette smoking.

Results: Regular opium consumption was associated with an increased risk of BC (OR 3.5, 95% CI: 2.8, 4.3) compared with subjects who never used opium. Compared with continuous users, the risk decreased to one-third for those who stopped opium more than 10 years ago. The adjusted OR for those who used both crude opium (teriak) and opium juice was 7.4 (95% CI: 4.1, 13.3). There was a joint effect of opium and tobacco (OR for users of both opium and tobacco 7.7, 95% CI: 6.0, 9.7).

Conclusions: Regular opium use is associated with an approximately 4-fold risk for BC. The OR decreases along with the increasing time since stopping opium use.

Key words: IROPICAN, opium, bladder cancer

Introduction

Bladder cancer (BC) is the 10th most common type of cancer worldwide[1] and the 4th most common cancer among men in Iran,\\\ with an estimated age-standardized (World Standard) incidence rate (ASR) of 14.3/100 000 in 2020.[2] An increasing number of incident cases of BC is projected for Iran, due to ageing and population growth and lifestyle changes.[3] Tobacco use and occupational exposure to chemical substances (e.g. metalworking fluids, diesel exhaust, polycyclic aromatic hydrocarbons, benzidine[4]) are the most important risk factors for BC.[1,5]

Opium, a highly addictive substance obtained from the unripe seedpod of the poppy plant, is illicitly consumed by millions of people worldwide, particularly in the Middle East and South Asia.[6] Freshly taken from the poppy plant, opium contains alkaloids (e.g. morphine, codeine and thebaine). In these countries, it is often minimally processed by heating, boiling and drying and is variably adulterated with some

Key Messages

- To our knowledge, this is the first large-scale case study investigating the relationship between opium use and bladder cancer.
- There was a substantial decrease in bladder cancer risk after stopping opium use.
- There was an additive interaction between opium and tobacco use.

chemicals (e.g. lead or chromium) before it reaches consumers. In this minimally processed form, opium may be consumed as crude opium (teriak), opium sap (shireh) or opium dross (sukhteh).[7] These forms of opium may be ingested or smoked. Therefore, similar to tobacco, opium is a complex substance with many chemicals.

In recent systematic reviews,[8,9] opium use was suggested as a potential risk factor for BC. However, the risk of under-reporting and detection bias in these studies was high.[8,9] Moreover, some of the included studies suffered from methodological limitations, including lack of controlling for confounding variables (age, sex, cigarette smoking), small samples size and lack of information about starting age of opium use, duration of use, dose and route of consumption. A recent study about the association between opium use and BC in one of the provinces in Iran suggested that regular use of opium had been more common among BC patients than among people in their neighbourhood.[10] None of the previous studies investigated the effect of time since stopping opium use, nor the dose-response relationship between opium use and BC risk. An International Agency for Research on Cancer (IARC) Working Group in 2020 concluded that opium use has a carcinogenic effect on humans, based on sufficient evidence of carcinogenicity in humans.[11] BC is one of the cancers which has been shown to have a positive association with opium consumption.

In the present large-scale study, we report the association between various parameters of opium use and the risk of BC.

Methods

The IROPICAN case-referent study was launched in 10 provinces in Iran to assess the association between opium consumption and risk of cancers of the lung, colorectum, bladder, and head and neck, compared with a joint group of referents who were frequency-matched by gender, age and place of reference with cancer cases of all four cancer types combined. These provinces were selected because the prevalence of opium use was relatively high, and also access to referral hospitals was possible. The referents were enrolled concurrently with the cases among the relatives or friends of patients from non-oncology wards or others who visited the hospital for reasons other than receiving treatment. The referents had to be free of cancer at the date of recruitment. Details of the study have been described elsewhere.[12]

For the current study, we use data of histologically confirmed primary BCs (ICD-O: C67) admitted to referral hospitals, who were recruited as cases from May 2017 until July 2020,[12] and the pool of all referents of the IROPICAN study. The mean age at recruitment was 63.6 years for the cases and 57.4 years for the referents. All BCs were incident cases diagnosed less than 1 year before the interview.

Altogether 717 BC cases and 3477 referents were recruited to the study. Out of the cases, 587 (81.9%) were urothelial carcinomas and 130 (18.1%) were BC cases of other and unknown histology. The characteristics of the cases and controls are given in Table 1. The non-response rate among the cases was 1% and among the referents 11%, with the main reasons for non-participation including sickness and lethargy among cancer patients and lack of time or unwillingness to donate a biological sample among referents.[12]

Exposure assessment

Detailed histories of opium use among both cases and referents were collected, including duration of use, starting and stopping ages, amount, and frequency of opium use per day, week and month. The amount of opium use was asked in local units of opium use and converted to grams. Other information collected included type of opium (crude opium, opium juice and both types) and routes of administration (only smoking, only ingestion and both routes). Information on the amount of opium use at a time (in grams) and number of times per day (frequency) was also collected. All these metrics were answered for up to five separate periods of opium use, and the durations of these periods were used as weights in the calculation of weighted averages.

In the statistical analyses, ever-use of opium was defined as using opium at least once during a lifetime, and regular

Table 1 Distribution of demographic characteristics and habits of the bladder cancer cases and referents at the time of interview in Iran from May 2017 to July 2020

Variable	Bladder cancer cases Number (%)	Referents Number (%)
Total	717 (100)	3477 (100)
Age		
30–39	14 (2.0)	257 (7.4)
40–49	50 (7.0)	559 (16.1)
50–59	181 (25.2)	1070 (30.8)
60–69	267 (37.2)	1092 (31.4)
≥70	205 (28.6)	499 (14.4)
Gender		
Female	93 (13.0)	1077 (31.0)
Male	624 (87.0)	2400 (69.0)
Place of residence		
Capital city of the region	267 (37.2)	1310 (37.7)
Other	450 (62.8)	2167 (62.3)
Province		
Tehran	139 (19.4)	816 (23.5)
Fars	166 (23.2)	943 (27.1)
Kerman	150 (20.9)	525 (15.1)
Golestan	46 (6.4)	374 (10.8)
Mazandaran	24 (3.4)	136 (3.9)
Kermanshah	52 (7.3)	251(7.2)
Khorasan-Razavi	30 (4.2)	170 (4.9)
Bushehr	56 (7.8)	84 (2.4)
Hormozgan	27 (3.8)	78 (2.2)
Systan-Balouchestan	27 (3.8)	100 (2.9)
Occupation		
High-skilled white-collar	202 (28.2)	1011 (29.1)
Low-skilled white-collar	153 (21.3)	575 (16.5)
High-skilled blue-collar	273 (38.1)	966 (27.8)
Low-skilled blue-collar	89 (12.4)	925 (26.6)
Cigarette smoking (pack-years)		
Non-smoker	287 (40.0)	2500 (71.9)
<15	111 (15.5)	449 (12.9)
15–31	120 (16.7)	255 (7.3)
>31	184 (25.7)	229 (6.6)
Unknown	15 (2.1)	44 (1.3)

opium use was defined as using opium at least once a week for at least 6 consecutive months. The cumulative amount of lifetime opium use was defined as the total duration of opium use (days) multiplied by the average daily amount, which was the product of an average amount of opium used at a time and the average daily frequency of opium use. We used a 3-year lag time, which means that opium consumption during the 3 past years before the interview date was excluded.

Ever-use of cigarettes and tobacco (waterpipe, Chopogh, Nass and pipe) was defined as using any at least once during a lifetime. Regular cigarette smoking and tobacco use were defined as using any at least once a week for at least 6 consecutive months. Also, cigarette smoking was defined as light (<14 pack-years), moderate (14–20) and heavy (>20). Furthermore, occupation was defined as high- or low-skilled white-collar, high- or low-skilled blue-collar.

Statistical analyses

Unconditional logistic regression models were used to measure adjusted odds ratios (OR) and 95% confidence intervals (CI). The ORs were adjusted for age, gender, province and pack-years of cigarette smoking. Occupation was dropped from the final models because this variable did not improve the model fit ($P>0.2$). In all analyses, non-users of opium were considered as the reference group. All statistical analyses were conducted using Stata, version 16 (Stata Corp., College Station, TX, licensed to Tampere University).

Results

Regular opium use was more than 3-fold among BC cases than among referents (adjusted OR 3.4, 95% CI: 2.7, 4.3; Table 2). The OR of regular opium use for bladder cancer of urothelial histology was 3.5 (95% CI: 2.7, 4.4) and for other or unknown histology 2.3 (95% CI: 1.2, 4.3).

The OR for those who used both teriak and shireh was 7.4 (95% CI: 4.1, 13.3) compared with non-users. Ingestion of opium was more strongly associated with an increased risk of BC than smoking of opium. Moreover, in a model adjusted with the duration of opium use, the OR for those who applied the ingestion route of opium use showed a strong association, with an OR of 6.8 (95% CI: 3.6, 13.6).

Those with a cumulative consumption of less than 4 kg opium during their life had a 2.3-fold risk of BC (95% CI: 1.7, 3.1), and the OR increased to 5.2 (95% CI: 3.7, 5.3) among those who had used more than a 16-kg cumulative amount of opium.

The duration of regular opium use was moderately associated with the risk of BC. The duration of fewer than 19 years showed an OR of 2.6 (95% CI: 1.9, 3.5) and the ORs for longer duration categories were ~4.5 (Table 2).

The average amount of opium used each time did not markedly affect the BC risk, but the frequency of daily opium use was highly associated with an increased risk of BC (Table 2). Those who used opium less than once per day had an OR of 2.1 (95% CI: 1.6, 2.8) whereas those who used opium more than two times per day had an OR of 9.5 (95% CI: 5.8, 15.4). This effect was also reflected in the OR for a lifelong cumulative count of opium use.

Table 2 Characteristics of opium use among regular opium users, and the odds ratios with opium use for bladder cancer in Iran from May 2017 to July 2020, from models including age, gender, province, cigarette pack-years. Lag 3 years

Metric of opium use	Bladder cancer cases Number (%)	Referents Number (%)	Adjusted odds ratio (95% CI)
Opium use			
Non-user	387 (54.0)	2881 (82.9)	Ref.
Irregular	27 (3.8)	135 (3.9)	1.1 (0.7, 1.8)
Regular[a]	303 (42.0)	461 (13.3)	3.4 (2.7, 4.3)
Type of opium used			
Crude opium (teriak)	251 (35.0)	405 (11.7)	3.2 (2.5, 4.0)
Opium juice (shireh)	20 (2.8)	32 (0.9)	3.8 (2.0, 7.0)
Both types	32 (4.5)	24 (0.7)	7.4 (4.1, 13.3)
Route of opium use			
Only smoking	209 (29.2)	383 (11.0)	2.8 (2.2, 3.6)
Only ingestion	27 (3.8)	30 (0.9)	6.3 (3.6 11.3)
Both routes	65 (9.1)	45 (1.3)	6.9 (4.5, 10.8)
Unknown	2 (0.3)	3 (0.1)	
Count per day[b]			
<1	141 (19.7)	345 (9.9)	2.1 (1.6, 2.8)
1–2	108 (15.1)	81 (2.3)	7.5 (5.3, 10.8)
>2	54 (7.5)	35 (1.0)	9.5 (5.8, 15.4)
Average of opium use at a time (g)			
<1	153 (21.3)	207 (6.0)	3.8 (2.9, 5.1)
1–2	62 (8.7)	146 (4.2)	2.3 (1.6, 3.3)
>2	88 (12.3)	108 (3.1)	4.1 (2.9, 5.9)
Daily dose of opium (g)[c]			
<1	141 (19.7)	268 (7.7)	2.7 (2.1, 3.6)
1–2	54 (7.5)	84 (2.4)	3.5 (2.3, 5.2)
>2	108 (15.1)	109 (3.1)	5.3 (3.8, 7.4)
Starting age of opium use			
<20	21 (2.9)	53 (1.5)	2.5 (1.4, 4.5)
20–29	89 (12.4)	143 (4.1)	3.3 (2.4, 4.7)
30–39	105 (14.6)	112 (3.2)	5.1 (3.6, 7.1)
≥40	88 (12.3)	153 (4.4)	2.8 (2.0, 3.8)
Time since stopping opium use (years)			
Current user	197 (27.5)	223 (6.4)	4.8 (3.7, 6.3)
<10	74 (10.3)	138 (4.0)	3.0 (2.1, 4.2)
≥10	31 (4.3)	93 (2.7)	1.5 (1.0, 2.4)
Unknown	1 (0.1)	7 (0.2)	–
Duration of opium use (years)			
< 9	87 (12.1)	223 (6.4)	2.6 (1.9, 3.5)
19–29	106 (14.8)	131 (3.8)	4.5 (3.3, 6.2)
>29	110 (15.3)	107 (3.1)	3.8 (2.7, 5.3)
Cumulative amount of opium (kg)[d]			
<4	96 (13.4)	229 (6.6)	2.3 (1.7, 3.1)
4–14	87 (12.1)	115 (3.3)	4.1 (2.9, 5.8)
>14	117 (16.3)	114 (3.3)	5.2 (3.7, 7.2)
Unknown	3 (0.4)	3 (0.4)	–

(Continued)

Table 2 Continued

Metric of opium use	Bladder cancer cases Number (%)	Referents Number (%)	Adjusted odds ratio (95% CI)
Cumulative count of opium use (times)[e]			
<4900	64 (8.9)	230 (6.6)	1.6 (1.1, 2.2)
4900–11 000	77 (10.7)	116 (3.3)	3.8 (2.7, 5.4)
>11000	161 (22.5)	114 (3.3)	6.8 (5.0, 9.3)
Unknown	1 (0.1)	1 (0.0)	–

[a]Regular opium use: using opium at least once a week for at least a 6-month consecutive period during the lifetime.
[b]Duration-weighted average of the period-specific daily frequencies of opium use.
[c]Count per day multiplied by the average of opium use at a time (g).
[d]Cumulative amount: the average daily amount of opium multiplied by the total duration of opium use (days).
[e]Cumulative count: average count per day multiplied by the total duration of opium use (days).

Those who had used opium more than 11 000 times had an OR of 6.8 (95% CI: 5.0, 9.3).

The OR did not have a consistent association with the age of starting opium use. The risk was highest among those who started at the age of 30–39 years (Table 2). The risk of BC among current opium users was 4.8 (95% CI: 3.7, 6.3) but the OR dropped to 1.5 (95% CI: 1.0, 2.4) among those who had stopped opium use more than 10 years before the date of interview (Table 2). In a model adjusted with cumulative opium use after further adjustment of the previous analysis for the cumulative amount of opium use, the OR for those who had stopped opium use more than 10 years before the interview date was 0.3 (95% CI: 0.2, 0.4) and for those who had stopped opium use less than 10 years before index date was 0.5 (95% CI: 0.4, 0.7) as compared with those who still used opium at the index date (results not shown in the tables).

In the model including age, gender, province and opium use, the OR for cigarette smokers with less than 14 pack-years was 1.8 (95% CI: 1.4, 2.4), for those with 14–20 pack-years was 2.9 (95% CI: 2.2, 3.8) and for those with more than 20 pack-years was 1.2 (95% CI: 0.5, 2.6), compared with non-smokers.

The results presented above and shown in Table 2 are for males and females combined. The OR for regular opium use among females was 2.9 (95% CI: 1.0, 8.2) and among males 3.4 (95% CI: 2.7, 4.4). Because there were only 93 BC cases among women, the data do not allow study of the effects of specific measures of opium use for women.

The adjusted OR for those who used both opium and tobacco was 7.7 (95% CI: 6.0, 9.7), as compared with those who did not use tobacco or opium (Table 3). When the analysis was restricted to smoking opium only, the respective OR was 7.4 (95% CI: 5.6, 9.7).

Discussion

In this large multicentre case-referent study, regular opium use was associated with an approximately 4-fold risk for BC compared with the subjects who never used opium. The OR was similar for those with confirmed urothelial histology and for those with other or unknown histology, and similar for males and females. Those who used both crude opium and opium juice had a 7-fold risk of BC. Ingested opium carried a higher risk of BC than smoked opium. The risk also increased if the duration of opium was more than 17 years or cumulative use was more than 4 kg. The risk increased along with increasing frequency of daily usage, but the average amount of opium used each time did not have much effect. The starting age of opium use did not have a major independent role in the BC risk.

Table 3 Odds ratios of the interaction of regular opium use and tobacco use (cigarette, water pipe, pipe, chewing tobacco, Chopogh) to the risk of bladder cancer in Iran from May 2017 to July 2020, adjusted for age, gender, and province. Lag 3 years

Tobacco use[a]	Opium use[a]			
	Never		Regular	
	Cases/referents	OR (95% CI)	Cases/referents	OR (95% CI)
Never	171/1951	Ref.	40/101	3.8 (2.5, 5.7)
Regular	197/725	2.2 (1.7, 2.7)	259/349	7.7 (6.0, 9.7)[b]

[a]Results for irregular opium use (28 cases, 135 referents) and irregular tobacco use (23 cases, 228 referents) not shown.
[b]Relative excess risk due to interaction: 2.7 (95% CI: 0.7, 4.7), attributable proportion due to interaction 0.4 (0.1, 0.6), synergy index 1.7 (1.1, 2.6).

Few studies, mostly consisting of small-sized case-referent studies, have evaluated the effect of opium use on the risk of BC.[8,11,13–15] Our results showed that opium consumption increases the risk of BC by 3-fold as compared with those who have not used opium (OR: 3.4, 95% CI: 2.7, 4.3), which concurs with results of previous case-control studies on opium and BC. Ghadimi *et al.*[14] showed that opium use was associated with an increased risk of BC with an OR of 5.0 (95% CI: 1.1, 2.3). Akbari *et al.*[15] reported an OR of 3.9 (95% CI: 1.2, 12). A systematic review also showed that opium use was associated with an increased risk of BC compared with non-users, with a pooled OR of 3.9 (95% CI: 3.1, 4.9).[8] Another systematic review of opium as a carcinogen showed that the pooled OR based on fixed effect model analysis was 4.1 (95% CI: 3.2, 5.1) and based on random effect model analysis was 3.8 (95% CI: 2.7, 5.4).[9] A recent case-control study from Kerman province in Iran compared opium use in BC patients diagnosed 2013–15—i.e. slightly earlier than the cases of our study (2016–20)—with neighbourhood controls, and observed an OR of 4.4 (95% CI: 2.9, 6.5) for regular opium use, which is similar to the OR seen in our study despite differences in control selection and analysis methods.[10] Our result restricted to urothelial BC is in line with the study conducted by Zeighami *et al.*[16] who showed that ever-use of opium use was more common among urothelial BC cases than among referents (OR: 3.0, 95% CI: 1.6, 5.4).

The carcinogenicity mechanism of opium is not completely clear. The IARC working group found strong evidence that opium dross and opium pyrolysates exhibit characteristics of carcinogens.[11] Another explanation is that opium use promotes tumorigenesis by influencing angiogenesis and immunosuppression and by facilitating cancer cell proliferation.[17–19] Furthermore, the exposure of the bladder to carcinogens will increase because alkaloids in opium cause urinary retention and cystitis.[20]

Our study suggested that opium consumption by smoking carried a lower risk of BC than the ingestion route. Additionally, after considering the duration of opium use, the ingestion route still showed a higher risk of BC, which is in line with the study by Sheikh *et al.*[13] which showed an OR of 2.6 (95% CI: 1.2, 5.4) for the smoking route and 3.8 (95% CI: 1.6, 8.9) for the ingestion route of opium use.

A novel finding of this study was the strongly decreased risk of BC for those who had stopped opium use more than 10 years before the index date as compared with those who had used the same amount but had not stopped. To our knowledge, this is the first study reporting such an observation.

We observed an additive interaction effect between opium use and tobacco. Consistently with previous studies,[21,22] the risk of BC for tobacco alone was 2-fold but 7-fold for regular users of both opium and tobacco. There is one earlier study suggesting a joint effect of opium and cigarette smoking on the risk of BC.[22] In that study, the interaction was multiplicative but based on only one BC case who had used opium only.

It is possible that some persons have started opium use because of pain related to symptoms of BC. Because we used a 3-year lag period in our analyses, i.e. opium use during the last 3 years before the interview was not counted, our results should be free of reverse causality bias. Even without such lag assumption, the risk of reverse causality would be small because there were only 11 cases and 17 referents who started opium use less than 3 years before the interview. Most of the opium users among both cases and referents had been using opium for more than 20 years.

A major challenge in observational studies on the effect of opium use is to collect reliable data among both cases and referents, because opium use is a stigmatized and criminal offence. This might cause misclassification bias. However, it was shown in previous studies that the sensitivity of self-reporting of opium use among cases and referents was similar.[12,23]

Our study had several strengths such as a large sample size, histological confirmation for all BC cases, and use of healthy hospital visitor referents, unlike other hospital-based case-control studies in which the referents had other diseases.[23] The data quality in our study is high because data were collected by trained interviewers using a validated questionnaire.[12] Due to access to detailed data on the amount of opium over time, we were able to examine the dose-response association of opium use and BC as well the effects of timing of opium use. We also had detailed information on the main confounder, i.e. tobacco, which was included in the statistical models. The response rate among both BC patents and referents was high. Although we were able to control for several potential confounders, the effect of unknown or unmeasured confounders or the residual confounding of those measured cannot be neglected.

In conclusion, the risk of BC was higher among those who were regular opium users than among those who had never used opium, with evidence of a dose-response association with frequency and cumulative amount of use. The risk decreased after 10 years following stopping the use of opium. These results are in agreement with the IARC monograph volume 126, September 2020,[11] indicating a causal association between opium use on different types of cancers, including BC. Our study has important

implications for public health practice and policy making, not only in Iran but also among opium users in other countries.

Disclaimer

Where authors are identified as personnel of the International Agency for Research on Cancer/World Health Organization, the authors alone are responsible for the views expressed in this article and they do not necessarily represent the decisions, policy or views of the International Agency for Research on Cancer/World Health Organization.

Ethics approval

The study was approved by the Ethics Committee of the National Institute of Medical Research Development (NIMAD) (Code: IR.NIMAD.REC.1394.027). All participants signed written informed consent to participation in the study.

Data availability

The data underlying this article cannot be shared publicly due to privacy of individuals who participated in the study. Data may be shared on reasonable request to the corresponding author.

Author contributions

M.H., H.R., M.M., M.G.H. did the literature review. H.R., A.N., M.G.H., M.M., E.M., R.S., M.S., B.H., M.B., R.A., V.A., S.S.H., A.N., F.N., A.M., A.R. contributed to data collection. Also, F.Z., R.M., M.N. provided clinical consultation. H.P., A.R., R.M., P.B., E.W., F.K., K.Z., E.P. designed the study. M.H., E.P. did the data analysis, interpreted data and prepared the manuscript draft. All authors critically appraised the drafts of the manuscripts and approved the final version. K.Z. is the guarantor of the study and E.P. is the senior author of the manuscript.

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Conflict of interest

None declared.

References

1. Sung H, Ferlay J, Siegel RL *et al.* Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin* 2021;**71**: 209–49.
2. Roshandel G, Ghanbari-Motlagh A, Partovipour E *et al.* Cancer incidence in Iran in 2014: results of the Iranian national population-based cancer registry. *Cancer Epidemiol* 2019;**61**: 50–58.
3. Rafiemanesh H, Lotfi Z, Bakhtazad S *et al.* The epidemiological and histological trend of bladder cancer in Iran. *J Cancer Res Ther* 2018;**14**:532–36.
4. Cumberbatch MGK, Jubber I, Black PC *et al.* Epidemiology of bladder cancer: a systematic review and contemporary update of risk factors in 2018. *Eur Urol* 2018;**74**:784–95.
5. McGuire S. World Cancer Report 2014. Geneva, Switzerland: World Health Organization, International Agency for Research on Cancer, WHO Press, 2015. *Adv Nutr* 2016;**7**:418–19. 2016
6. United Nations. World Drug Report *2019*. New York, NY: UN Publication, 2019, sales no. E.19.XI, doi:10.1002/9781118929803.ewac0543.
7. Amin-Esmaeili M, Rahimi-Movaghar A, Sharifi V *et al.* Epidemiology of illicit drug use disorders in Iran: Prevalence, correlates, comorbidity and service utilization results from the Iranian mental health survey. *Addiction* 2016;**111**:1836–47.
8. Afshari M, Janbabaei G, Bahrami MA *et al.* Opium and bladder cancer: A systematic review and meta-analysis of the odds ratios for opium use and the risk of bladder cancer. *PLoS ONE* 2017; **12**:e0178527.
9. Bidary MZ, Sahranavard M, Rezayat AA *et al.* Opium as a carcinogen: A systematic review and meta-analysis. *EClinical Medicine* 2021;**33**:100768.
10. Rashidian H, Haghdoost AA, Hadji M *et al.* Association between opium use and bladder cancer: A case-control study in a high risk area of Iran. *Clin Epidemiol Glob Health* 2021;**11**: 100772.
11. Warnakulasuriya S, Cronin-Fenton D, Jinot J *et al.* Carcinogenicity of opium consumption. *Lancet Oncol* 2020;**21**: 1407–08.
12. Hadji M, Rashidian H, Marzban M *et al.* The Iranian study of opium and cancer (IROPICAN): rationale, design, and initial findings. *Arch Iran Med* 2021;**24**:167–76.
13. Sheikh M, Shakeri R, Poustchi H *et al.* Opium use and subsequent incidence of cancer: Results from the Golestan cohort study. *Lancet Glob Health* 2020;**8**:649–60.
14. Ghadimi T, Gheitasi B, Nili S *et al.* Occupation, smoking, opium, and bladder cancer: A case–control study. *South Asian J Cancer* 2015;**4**:111–14.
15. Akbari M, Naghibzadeh TA, Khanjani N *et al.* Opium as a risk factor for bladder cancer: a population-based case-control study in Iran. *Arch Iran Med* 2015;**18**:567–71.
16. Zeighami S, Azizzadeh E, Tabatabaee HR, Adib A, Babaei AH, Ariafar A. Opium and grade of urothelial bladder cancer. *J Nephropathol* 2017;**7**:69–73.
17. Vallejo R, de Leon-Casasola O, Benyamin R. Opioid therapy and immunosuppression: a review. *Am J Ther* 2004;**11**:354–65.
18. Grandhi RK, Lee S, Abd-Elsayed A. Does opioid use cause angiogenesis and metastasis? *Pain Med* 2017;**18**:140–51.

19. Sheikh M, Kamangar F, Malekzadeh R. Fifty years of research and one conclusion: opium causes cancer. *Arch Iran Med* 2020; **23**:757–60.
20. Kamangar F, Shakeri R, Malekzadeh R. Opium use: an emerging risk factor for cancer? *Lancet Oncol* 2014;**15**:69–77.
21. Cumberbatch MGK, Jubber I, Black PC *et al.* Epidemiology of bladder cancer: a systematic review and contemporary update of risk factors in 2018. *Eur Urol* 2018;**74**:784–95.
22. Sadeghi A, Behmard S, Vesselinovitch SD. Opium: a potential urinary bladder carcinogen in man. *Cancer* 1979;**43**:2315–21.
23. Rashidian H, Hadji M, Marzban M *et al.* Sensitivity of self-reported opioid use in case-control studies: Healthy individuals versus hospitalized patients. *PLoS One* 2017;**12**: e0183017.

PUBLICATION IV

Opium Use and Risk of colorectal cancer: A Multi-center Case-Referent Study in Iran

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ORIGINAL ARTICLE

Opium use and risk of colorectal cancer: a multi-center case-referent study in Iran

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ABSTRACT

Background: Opium use has been associated with an increased risk of cancers of the lung, oesophagus, and pancreas, and it was recently classified by the International Agency for Cancer Research as carcinogenic to humans. It is not clear whether opium also increases the risk of colorectal cancer (CRC). The aim of our study was to assess the association between various metrics of opium use and the risk of CRC.

Methods: This case-referent study from seven provinces in Iran comprised 848 CRC cases and 3215 referents. Data on opium use (duration, amount, frequency) and potential confounders were collected by trained interviewers. Multivariable unconditional logistic regression models were used to measure odds ratios (OR) adjusted for age, gender, province, marital status, family history of CRC-linked cancers, consumption of red meat, fruits and vegetables, body shape, occupational physical activity, and socioeconomic status.

Results: Regular opium consumption was not associated with the risk of CRC (OR 0.9, 95% confidence interval, CI: 0.7, 1.2) compared to subjects who never used opium. However, frequent opium use more than twice a day was associated with an increased risk of CRC compared to non-users of opium (OR: 2.0, 95% CI: 1.1, 3.8; p for quadratic trend 0.008).

Conclusion: There seems to be no overall association between opium use and CRC, but the risk of CRC might be increased among persons who use opium many times a day.

Abbreviations: CRC: Colorectal cancer; SES: Socioeconomic status; OR: Odds ratios; CI: Confidence interval; FINJEM: Finnish job exposure matrix; PPWL: Physical activity workload

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Background

Colorectal cancer (CRC) is the third most common cancer and second most deadly malignancy, with 1.9 million incident cases and 0.9 million death worldwide in 2020 [1–3]. There were 1.2 million incident cases of colon cancers and 0.7 million rectal cancers worldwide in 2020, and the numbers are projected to increase until 2040 [1,2], mainly due to the increasing number of older persons [2]. Risk factors of CRC include age, obesity, lack of physical activity, red meat intake, and constipation [4]. Opium use causes constipation [5], and an excess risk of CRC among opium users is plausible through this mechanism. In Iran in 2014, CRC was the third most common cancer among men with an estimated age-standardized (World Standard) incidence rate (ASR) of 16.6/100 000 and the second most common cancer among women (ASR: 11.9/100 000) [6].

Opium, a highly addictive substance obtained from the unripe seedpod of the poppy plant, is illicitly consumed by millions of people worldwide, particularly in the Middle East and South Asia [7]. Freshly taken from the poppy plant, opium contains alkaloids (e.g., morphine, codeine, and thebaine). It is often minimally processed by heating, boiling, drying, and variably adulterated with some chemicals (e.g., lead or chromium) before it reaches consumers. In this minimally processed form, opium may be consumed as crude opium (teriak), opium juice (shireh), or opium dross (sukhteh) [8]. These forms of opium may be ingested or smoked.

Opium usually contains lead as an additive [9–12], and lead can cause constipation [13]. Since risk of CRC is elevated among those who suffer from constipation [13], we hypothesized that constipation might have an intermediate role between opium consumption and risk of CRC. Another mediator between opium use and risk of CRC may be poor oral hygiene. Opium use is associated with periodontal disease, and poor oral hygiene might increase risk of CRC [14].

An IARC Working Group in 2020 concluded that opium use has a carcinogenic effect on humans, based on sufficient evidence of carcinogenicity in humans for cancers of the lung, oesophagus, and pancreas [15]. The association between opium use and CRC is less clear. In two recent studies, opium use was suggested as a potential risk factor for CRC [16,17] while one study showed no association [18]. These studies suffered from limitations, such as small numbers of cases, reporting bias, lack of controlling for confounding variables, and lack of detailed information on opium use. In the present large-scale study, we report the association between various metrics of opium use and the risk of CRC.

Material and methods

The IROPICAN case-referent study was launched in ten provinces in Iran to assess associations between opium consumption and risk of cancers of the lung, CRC, bladder, and head and neck. These provinces were selected because the prevalence of opium use was relatively high and access to referral hospitals was possible. In this study we use data from only seven provinces since the data collection of CRCs was not performed in three centers in the southern part of Iran.

In the IROPICAN study, referents were enrolled concurrently with the cases among the relatives or friends of patients from non-oncology wards or others who visited the hospital for reasons other than receiving treatment. They were frequency-matched by gender, age, and province of residence, with cancer cases of all four cancer types combined. The referents had to be free of cancer at the date of recruitment. Trained interviewers conducted face to face interviews and collected data using a comprehensive validated questionnaire and 12 ml of venous whole blood for all cancer cases and referents. Details of the study have been described elsewhere [19].

For the current study, we used data of histologically confirmed incident primary CRCs (ICD-O-3: C18, colon; C19-20, rectum) and referents recruited from May 2017 until July 2020 [19]. The non-response rate among the cases was 1% and among the referents 11%. The main reasons for non-participation among the cancer patients were sickness and lethargy, and among the referents lack of time or unwillingness to donate a biological sample [19]. Forty cases with unknown subsite were excluded from the analysis. Altogether 848 CRC cases and 3215 referents were included in the study. The mean age at recruitment for the included cases was 58.7 (standard deviation 12.5) years and for the referents 57.2 (standard deviation 11.5) years.

Exposure assessment

Detailed histories of opium use were collected from cases and referents, including types of opium (crude opium, opium juice, and both types), routes of administration (only smoking, only ingestion, and both routes), duration of use, starting and stopping age, and daily amount and frequency of opium use. The amount of opium use was asked in local units of opium use which were converted to grams. All these metrics were answered for up to 5 separate periods of opium use, and the durations of these periods were used as weights in the calculation of weighted averages of the count per day (frequency), average amount of opium used at a time and daily dose of opium use.

In the statistical analyses, ever-use of opium was defined as using opium at least once during a lifetime, and regular opium use was defined as using opium at least once a week for at least six consecutive months. The cumulative amount of lifetime opium use was defined as the total duration of opium use (days) multiplied by the average daily amount, which was the product of an average amount of opium used at a time and the average daily frequency of opium use. We used a three-year lag time which means that possible opium consumption during the three last years before the interview date was excluded.

Potential confounders

Those who reported one or more of cancers of the colon, rectum, stomach, ovary and endometrium in their

Table 1. Numbers (and percentages) of subjects of colorectal cancer (CRC) cases and referents, by demographic characteristics and life habits.

Variable category	CRC cases	Colon cancer cases	Rectal cancer cases	Referents
Total	848 (100)	455 (100)	393 (100)	3215 (100)
Age at interview				
30-39	66 (7.8)	34 (7.5)	32 (8.1)	250 (7.8)
40-49	123 (14.5)	65 (14.3)	58 (14.8)	505 (15.7)
50-59	235 (27.7)	118 (25.9)	117 (29.8)	997 (31.0)
60-69	252 (29.7)	144 (31.7)	108 (27.5)	1020 (31.7)
≥70	172 (20.3)	94 (20.7)	78 (19.9)	443 (13.8)
Gender				
Female	366 (43.2)	198 (43.5)	168 (42.8)	1010 (31.4)
Male	482 (56.8)	257 (56.5)	225 (57.3)	2205 (68.6)
Place of residence				
Capital city of the region	538 (63.4)	300 (65.9)	238 (60.6)	1254 (39.0)
Other	310 (36.6)	155 (34.1)	155 (39.4)	1961 (61.0)
Province				
Tehran	170 (20.0)	100 (22.0)	70 (17.8)	816 (25.4)
Fars	248 (29.3)	102 (22.4)	146 (37.2)	943 (29.3)
Kerman	104 (12.3)	51 (11.2)	53 (13.5)	525 (16.3)
Golestan	140 (16.5)	92 (20.2)	48 (12.2)	374 (11.6)
Mazandaran	59 (7.0)	39 (8.6)	20 (5.1)	136 (4.2)
Kermanshah	66 (7.8)	32 (7.0)	34 (8.7)	251 (7.8)
Khorasan-Razavi	61 (7.2)	39 (8.6)	22 (5.6)	170 (5.3)
Socioeconomic status				
Low	325 (38.3)	173 (38.0)	152 (38.7)	863 (26.8)
Moderate	230 (27.1)	124 (27.3)	106 (27.0)	1085 (33.8)
High	293 (34.6)	158 (34.7)	135 (34.4)	1267 (39.4)
Body shape at age 15				
Normal	662 (78.1)	346 (76.0)	316 (80.4)	2694 (83.8)
Overweight	99 (11.7)	60 (13.2)	39 (9.9)	303 (9.4)
Obese	87 (10.3)	49 (10.8)	38 (9.7)	218 (6.8)
Physical activity at work				
Sedentary	282 (33.3)	157 (34.5)	125 (31.8)	1036 (32.2)
Medium	151 (17.8)	78 (17.1)	73 (18.6)	701 (21.8)
High	176 (20.8)	93 (20.4)	83 (21.1)	695 (21.6)
Unknown	239 (28.2)	127 (27.9)	112 (28.5)	783 (24.4)

first-degree relatives were defined as having a positive family history of cancer. These cancer types are possibly linked to Lynch syndrome or hereditary nonpolyposis colorectal cancer (HNPCC) [20,21].

We estimated perceived physical activity workload (PPWL) by using job histories of the participants and a Finnish job exposure matrix (FINJEM) [22,23]. In FINJEM there are two variables to estimate PPWL for each occupation, calendar period, the proportion of exposed (P), and mean level of the exposure among the exposed (L) [24]. We were not able to calculate the cumulative PPWL for the entire work life because of overlapping work periods in the collected data. Therefore, we only used PL for the longest work period, categorized into three categories: sedentary, moderate, or heavy (highest tertile of non-sedentary observations).

Numerous variables related to socioeconomic status (SES) were summarized using principal component analysis by combining data on years of education, and ownership of vacuum cleaner, dress washing machine, dishwashing machine, freezer, microwave, split air conditioner, laptop, internet access, mobile phone, personal car, owned house, or a shop. The weighted sums of these variables, where the weights are equal to the principal component loading, was categorized into tertiles and used in the logistic regression analyses as the SES variables.

Information about intake of 131 food items before the recruitment date was collected using a validated Persian cohort food frequency questionnaire [25]. For each food item, the reported daily frequency of consumption was multiplied by the standard portion size (grams). Consumption of red meat, fruit and vegetables was categorized into tertiles. Since we did not have information about height and weight in relevant ages to calculate body mass index, we classified body shape of the participants at age 15 into three categories (normal, overweight, obese) using data collected as pictograms which have been shown as a valid tool for such classification [26].

To define the constipation, participants were asked whether they have had constipation for more than six months. Number of decayed teeth was examined by trained interviewers and used as the index of oral hygiene, categorized based on the median (< 7, ≥7).

Statistical analyses

Unconditional logistic regression models were used to measure odds ratios (OR) and 95% confidence intervals (CI). The ORs were adjusted for age (ten-year age category), gender, and province, marital status, SES, body shape at age 15 years old, PPWL, family history of cancer, and intake of red meat and vegetables. Pack-year cigarette smoking, and energy intake were dropped from the final models because these variables did not significantly improve the model fit. In all analyses, non-users of opium were considered as the reference group.

To understand the existence and effect of the factors acting as possible mediators between opium and CRC, i.e.,

Table 2. Odds ratios (or) and 95% confidence intervals (CIs) for opium use, marital status, family history of cancer, red meat, vegetables, body shape, socioeconomic status, and perceived physical workload in risk of colorectal cancer, adjusted for age, gender, province, and all other cofactors listed in this table.

		Colorectal cancer		Colon cancer		Rectal cancer	
Variable category	Referents N (%)	N (%)	OR (95% CI)	N (%)	OR (95% CI)	N (%)	OR (95% CI)
Total	3215 (100)	848 (100)		455 (100)		393 (100)	
Opium use							
Non-user	2649 (82.4)	719 (84.8)	Ref.	386 (84.8)	Ref.	333 (84.7)	Ref.
Irregular	127 (3.9)	40 (4.7)	1.3 (0.9, 1.9)	25 (5.5)	1.4 (0.9, 2.3)	15 (3.8)	1.1 (0.6, 2.0)
Regular[a]	439 (13.7)	89 (10.5)	0.9 (0.7, 1.2)	44 (10)	0.8 (0.6, 1.2)	45 (11.5)	1.0 (0.7, 1.4)
Cofactors included in the model							
Marital status							
Married	2913 (90.6)	701 (82.7)	Ref.	374 (82.2)	Ref.	327 (83.2)	Ref.
Widow	164 (5.1)	99 (11.7)	1.9 (1.4, 2.6)	56 (12.3)	2.1 (1.4, 3.0)	43 (10.9)	1.8 (1.2, 2.6)
Divorced/separated	39 (1.2)	21 (2.5)	2.0 (1.1, 3.4)	10 (2.2)	1.8 (0.9, 3.9)	11 (2.8)	2.2 (1.0, 4.4)
Single	90 (2.8)	21 (2.5)	0.8 (0.5, 1.4)	12 (2.6)	1.0 (0.5, 1.8)	9 (2.3)	0.7 (0.4, 1.6)
Unknown	9 (0.3)	6 (0.7)	–	3 (0.7)	–	3 (0.8)	–
Socioeconomic status							
Low	863 (26.8)	325 (38.3)	Ref.	173 (38)	Ref.	152 (38.7)	Ref.
Moderate	1085 (33.8)	230 (27.1)	0.7 (0.5, 0.8)	124 (27.3)	0.7 (0.5, 0.9)	106 (27.0)	0.6 (0.5, 0.8)
High	1267 (39.4)	293 (34.6)	0.7 (0.6, 0.9)	158 (34.7)	0.8 (0.6, 1.0)	135 (34.4)	0.7 (0.5, 0.9)
Body shape at age 15							
Normal	2694 (83.8)	662 (78.1)	Ref.	346 (76)	Ref.	316 (80.4)	Ref.
Overweight	303 (9.4)	99 (11.7)	1.1 (0.9, 1.5)	60 (13.2)	1.3 (0.9, 1.8)	39 (9.9)	1.0 (0.7, 1.4)
Obese	218 (6.8)	87 (10.3)	1.3 (1.0, 1.8)	49 (10.8)	1.4 (1.0, 2.0)	38 (9.7)	1.3 (0.9, 1.9)
Perceived physical workload							
Sedentary	1036 (32.2)	282 (33.3)	Ref.	157 (34.5)	Ref.	125 (31.8)	Ref.
Medium	701 (21.8)	151 (17.8)	0.8 (0.6, 0.9)	78 (17.1)	0.7 (0.5, 1.0)	73 (18.6)	0.8 (0.6, 1.1)
High	695 (21.6)	176 (20.8)	0.8 (0.6, 1.0)	93 (20.4)	0.8 (0.6, 1.0)	83 (21.1)	0.8 (0.6, 1.1)
Unknown	695 (21.6)	239 (28.2)	0.6 (0.5, 0.8)	127 (27.9)	0.6 (0.4, 0.9)	112 (28.5)	0.6 (0.4, 0.9)
Family history of cancer							
No	3060 (95.2)	781 (92.1)	Ref.	419 (92.1)	Ref.	362 (92.1)	Ref.
Yes	155 (4.8)	67 (7.9)	1.7 (1.3, 2.3)	36 (7.9)	1.7 (1.2, 2.6)	31 (7.9)	1.7 (1.1, 2.6)
Red meat (g/day)							
< 12.83	1742 (54.2)	410 (48.4)	Ref.	215 (47.3)	Ref.	195 (49.6)	Ref.
12.83-25.64	990 (30.8)	233 (27.5)	1.3 (1.0, 1.5)	119 (26.2)	1.3 (1.0, 1.6)	114 (29.0)	1.3 (1.0, 1.7)
>25.64	480 (14.9)	205 (24.2)	2.2 (1.8, 2.7)	121 (26.6)	2.5 (1.9, 3.3)	84 (21.4)	1.9 (1.4, 2.6)
Unknown	3 (0.01)	–		–	–	–	–
Fruit and vegetables (g/day)							
<422	1606 (50.0)	409 (48.2)	Ref.	211 (46.4)	Ref.	198 (50.4)	Ref.
422-576	803 (25.0)	200 (23.6)	1.0 (0.8, 1.3)	108 (23.7)	1.2 (0.9, 1.5)	92 (23.4)	0.9 (0.7, 1.2)
>576	803 (25.0)	239 (28.2)	1.1 (0.9, 1.3)	136 (29.9)	1.3 (1.0, 1.7)	103 (26.2)	0.9 (0.7, 1.2)
Unknown	3 (0.01)	–		–	–	–	–

[a]Using opium at least once a week for at least six consecutive months during the lifetime. Some of them may have stopped opium use before the date of interview.

constipation and decayed teeth as an indicator of poor oral hygiene [27], we conducted a series of mediation analyses [28].

All statistical analyses were conducted using Stata, version 17 (Stata Corp, College Station, Texas 77845 USA, licensed to Tampere University).

Results

In the final study series, there were 455 cases of colon cancer and 393 cases of rectal cancer (Table 1). Out of 848 cases 89 (10.5%) and out of the 3215 referents 439 (13.7%) were regular opium users (Table 2).

Irregular opium use showed a non-significant 30% excess of CRC (Table 2). Regular opium use showed no association with risk of either CRC overall (OR: 0.9, 95% CI: 0.7, 1.2) or any of its two subsites (Table 2).

In analyses focusing to characteristics of opium use, ingestion of opium seemed to be associated with risk of rectal cancer (OR: 2.3, 95% CI: 1.0, 5.6) but not with colon cancer. The amount of opium used each time seemed not to affect risk of CRC (Table 3).

The frequency of opium use (count per day) was associated with an increasing risk of CRC (Table 3). Those who used opium more than two times per day had an OR of 2.0 (95% CI: 1.1, 3.8) compared to non-users of opium. Use of opium less than once per day showed an OR of 0.7 (95% CI: 0.5, 1.0). The quadratic p-trend related to the frequency of opium use for CRC was 0.008 (for colon 0.02, for rectum 0.1).

Among the cofactors included in the final model, red meat intake showed a strong association with the risk of CRC, more strongly for colon than rectum (Table 2). Obese persons showed a 30% higher risk of CRC than those with normal body shape. The risk of CRC was 70% higher among those who had first-degree relatives with HNPCC cancers than those without.

The mediation analysis showed that constipation and oral hygiene are mediators. However, the effect of opium use mediated *via* these factors was not strong, and hence the mediation analysis did not provide additional insight on the association between opium and risk of CRC. Therefore, we did not include constipation and oral hygiene in the final model.

Table 3. Odds ratios (or) and 95% confidence intervals (CIs) by characteristics of regular opium use from models adjusted for age, gender, province, marital status, family history of cancer, red meat, vegetables, body shape, socioeconomic status, and perceived p physical workload. Lag 3 years.

		Colorectal cancer		Colon cancer		Rectal cancer	
Metric of opium use category	Referents N (%)	N (%)	OR (95% CI)	N (%)	OR (95% CI)	N (%)	OR (95%CI)
Type of opium used							
No opium use	2649 (82.4)	719 (84.8)	Ref.	386 (84.8)	Ref.	333 (84.7)	Ref.
Crude opium (Teriak)	387 (12.0)	72 (8.5)	0.8 (0.6, 1.1)	34 (7.5)	0.8 (0.5, 1.2)	35 (8.9)	0.9 (0.6, 1.3)
Opium juice (Shireh)	31 (1.0)	9 (1.1)	1.3 (0.6, 2.8)	5 (1.1)	0.9 (0.3, 2.8)	7 (1.2)	1.6 (0.6, 4.4)
Both types	21 (0.7)	8 (0.9)	1.6 (0.7, 3.6)	5 (1.1)	1.6 (0.5, 4.9)	3 (0.8)	1.5 (0.5, 4.5)
Route of opium use							
Only smoking	368 (11.5)	69 (8.1)	0.9 (0.6, 1.2)	34 (7.5)	0.8 (0.6, 1.2)	35 (8.9)	0.9 (0.6, 1.3)
Only ingestion	27 (0.8)	12 (1.4)	1.6 (0.8, 3.2)	5 (1.1)	1.0 (0.4, 2.8)	7 (1.8)	2.3 (1.0, 5.6)
Both routes	41 (1.3)	8 (0.9)	0.9 (0.4, 2.0)	5 (1.1)	1.0 (0.4, 2.6)	3 (0.8)	0.8 (0.2, 2.7)
Unknown	3 (0.1)	–	–	–	–	–	–
Count per day[a]							
<1	308 (9.6)	48 (5.7)	0.7 (0.5, 1.0)	23 (5.1)	0.6 (0.4, 1.0)	25 (6.4)	0.8 (0.5, 1.2)
1-2	97 (3.0)	24 (2.8)	1.1 (0.7, 1.8)	12 (2.6)	1.1 (0.6, 2.1)	12 (3.0)	1.2 (0.6, 2.3)
>2	34 (1.1)	17 (2.0)	2.0 (1.1, 3.8)	9 (2)	2.0 (1.0, 4.5)	8 (2.0)	2.0 (0.9, 4.4)
Average opium uses at a time (g)							
<1	190 (5.9)	54 (6.4)	1.2 (0.9, 1.9)	28 (6.2)	1.1 (0.7, 1.8)	26 (6.6)	1.4 (0.9, 2.1)
1-2	140 (4.4)	18 (2.1)	0.6 (0.4, 1.0)	7 (1.5)	0.4 (0.2, 1.0)	11 (2.8)	0.7 (0.4, 1.4)
>2	109 (3.4)	17 (2.0)	0.7 (0.4, 1.2)	9 (2)	0.8 (0.4, 1.6)	8 (2.0)	0.7 (0.3, 1.4)
Daily dose of opium (g)[b]							
<1	233 (7.3)	46 (5.4)	0.9 (0.6, 1.2)	23 (5.1)	0.8 (0.8, 1.3)	23 (5.9)	1.0 (0.6, 1.6)
1-2	84 (2.6)	17 (2.0)	0.9 (0.5, 1.6)	9 (2)	0.9 (0.4, 1.9)	8 (2.0)	0.9 (0.4, 1.8)
>2	122 (3.8)	26 (3.1)	1.0 (0.6, 1.5)	12 (2.6)	0.9 (0.5, 1.8)	14 (3.6)	1.1 (0.6, 1.9)
Starting age of opium use							
<20	51 (1.6)	9 (1.1)	0.8 (0.4, 1.8)	5 (1.1)	0.9 (0.3, 2.3)	4 (1.0)	0.8 (0.3, 2.3)
20-29	134 (4.2)	20 (2.4)	0.7 (0.4, 1.2)	9 (2.0)	0.6 (0.3, 1.2)	11 (2.8)	0.9 (0.5, 1.7)
30-39	106 (3.3)	29 (3.4)	1.3 (0.8, 2.0)	16 (3.5)	1.3 (0.7, 2.3)	13 (3.3)	1.2 (0.7, 2.2)
≥40	148 (4.6)	31 (3.7)	0.8 (0.6, 1.3)	14 (3.1)	0.7 (0.4, 1.3)	17 (4.3)	1.0 (0.6, 1.7)
Time since stopping opium use							
Current user	213 (6.6)	41 (4.8)	0.9 (0.6, 1.3)	25 (5.5)	1.1 (0.9, 2.3)	16 (4.1)	0.8 (0.4, 1.3)
<10	133 (4.1)	31 (3.7)	1.0 (0.6, 1.5)	14 (3.1)	0.8 (0.5, 1.5)	17 (4.3)	1.2 (0.7, 2.0)
≥10	86 (2.7)	16 (1.9)	0.8 (0.5, 1.4)	5 (1.1)	0.5 (0.2, 1.3)	11 (2.8)	1.2 (0.6, 2.3)
Unknown	7 (0.2)	1 (0.1)	–	–	–	1 (0.3)	–
Duration of opium use (years)							
<19	254 (7.9)	60 (7.1)	1.0 (0.8, 1.4)	26 (5.7)	0.9 (0.6, 1.3)	34 (8.7)	1.2 (0.8, 1.8)
19-29	107 (3.3)	19 (2.2)	0.9 (0.5, 1.5)	13 (2.9)	1.2 (0.6, 2.2)	6 (1.5)	0.6 (0.3, 1.4)
>29	78 (2.4)	1 (1.2)	0.5 (0.3, 1.1)	5 (1.1)	0.5 (0.2, 1.2)	5 (1.3)	0.6 (0.2, 1.5)
Cumulative amount of opium (kg)[c]							
<4	211 (6.6)	45 (5.3)	0.9 (0.7, 1.3)	22 (2.2)	0.8 (0.5, 1.3)	23 (5.9)	1.1 (0.7, 1.7)
4 – 14	115 (3.6)	24 (2.8)	1.0 (0.6, 1.5)	10 (2.2)	0.8 (0.4, 1.5)	14 (3.6)	1.2 (0.6, 2.1)
>14	112 (3.5)	18 (2.1)	0.7 (0.4, 1.3)	11 (2.4)	1.0 (0.5, 1.7)	7 (1.8)	0.6 (0.3, 1.3)
Unknown	1 (0.0)	2 (0.2)	–	1(0.2)	–	1 (0.3)	–
Cumulative count of opium uses (times)[d]							
<4900	217 (6.8)	35 (4.1)	0.7 (0.5, 1.0)	17 (3.7)	0.7 (0.4, 1.1)	18 (4.6)	0.8 (0.5, 1.3)
4900-11000	126 (3.9)	30 (3.5)	1.1 (0.7, 1.8)	15 (3.3)	1.1 (0.6, 1.9)	15 (3.8)	1.2 (0.7, 2.2)
>11000	96 (3.0)	24 (2.8)	1.1 (0.7, 1.7)	12 (2.6)	1.0 (0.5, 1.8)	12 (3.1)	1.2 (0.6, 2.2)

[a]Duration-weighted average of the period-specific daily frequencies of opium use.
[b]Count per day multiplied by the average of opium use at a time (g).
[c]Cumulative amount: the average daily amount of opium multiplied by the total duration of opium use (days).
[d]Cumulative count: average count per day multiplied by the total duration of opium use (days).

Discussion

In this large multi-center case-referent study, regular opium use was generally not associated with risk of cancers of colon or rectum. Still, opium use of at least twice a day was associated with an approximately two-fold risk of CRC compared with the subjects who never used opium. The amount of opium used each time, or the duration and age span of opium use seemed not to affect the risk. Findings related to type and route of opium were also weak.

Few studies, two small case-referent studies and one cohort study, have evaluated the effect of opium use on the risk of CRC [16–18]. Our study showing lack of association with regular opium use is in line with the cohort study by Sheikh et al. [18]. In contrast, the two earlier case-referent studies showed a four-fold risk of CRC among opium users [16,17]. Both these studies used neighborhood referents. It has been reported that neighborhood controls may underestimate their opium use since using opium is not legal and users might not report it correctly [29]. In our study, instead, the referents were hospital visitors who were interviewed in identical way as the cases. It has been shown that in such a setting the completeness of reporting of opium use is similar among the cases and referents [30]. Still, there is chance of recall bias. The cases may report their opium use more likely than the referents. That would mean that our OR estimates are slightly too high.

To the best of our knowledge, our study is the first one that was able to evaluate the effect of frequency of opium use on the risk of CRC. In our data there was a trend of increasing CRC risk with increasing daily frequency of opium use, and use of opium more than twice a day carried a

two-fold risk of both cancers in colon and rectum. If these do not represent chance findings, we should seek for mechanisms how frequent opium use might increase risk of CRC. One possible mechanism considered *a priori* is constipation which can increase risk of CRC [13]. Lead as an additive of opium may lead to constipation [10]. A study by Nemati et al. showed that blood level of lead among opium users was three-fold as compared to non-users [31]. Although it is likely that frequent opium use increases risk of constipation, according to our data constipation appears not to have an important effect on the risk of CRC.

Heavy metals such as lead, arsenic, sulfate, chromium and cadmium as an opium additive [10,15] have been considered as risk factors for colon cancer; the plausible mechanism might be due to DNA damage along with dichromate ions to induce DNA methylation and gene silencing [32–34]. The study conducted by Sohrabi et al. showed that the mean concentration of lead in CRC tissues was approximately 2-fold as compared to that in healthy tissues [35]. The study by Etemadi et al. [36] showed that mortality and cancer risk might be increased due to presence of lead in the blood as a consequence of long term regular opium use. Our data did not include information on heavy metal intake of the cases and referents, and it was therefore not possible to assess direct effect of them on the CRC risk.

Because there is an association between frequency of opium use and risk of CRC, it would be expected that there also is an association between cumulative count of opium use and risk of CRC. We did however not observe any such association. The OR did not depend on the age of starting opium use nor on time since opium use had been stopped and increasing years of opium use rather decreased than increased the risk. It appears possible that opium users with constipation problems cannot continue opium use for a long time.

A study by Zu et al. [32] reported that opium use is associated with periodontal disease. Periodontal diseases might increase systemic inflammation, lead to immune system problems and alter gut microbiota, and probably influencing CRC carcinogenesis [14]. In our data number of decayed teeth used as an oral hygiene index did not show strong effect on the risk of CRC.

Another hypothetical mechanism that may explain the association of frequent opium use and increased risk of CRC could be related to polyps which are a confirmed risk factor of CRC [33,34]. If opium smoking increases risk of polyps in a similar way as has been shown for cigarette smoking [37] it would be possible that opium use may also indirectly increase the risk of CRC. We did not have data on polyps, but we suggest conducting studies on the effect of opium use on developing polyposis.

Our study had several strengths. The sample size was large, all CRC cases were histologically confirmed and correctly stratified as either colon or rectal cancer. We used healthy hospital visitor referents who do not share risk factors with the cases as might happen if the referents would be hospital patients with other diseases [30]. The data quality in our study was high because they were collected by trained interviewers using a validated questionnaire [19]. The response rate among both CRC patients and referents was high. Due to access to detailed information about details of opium use, we were able to examine the association of various metrics of opium use and risk of CRC. While accuracy of opium use information has been evaluated to be generally good, it has been shown that the assessment of amount of opium use is less reliable [38]. We do not know whether people who were asked to be referents differ in their opium consumption from the rest of the population. If they would be selected in terms of opium use, then we would have a selection bias. Therefore, we should be careful in interpreting the result in such a way that amount of opium use would have no effect.

In conclusion, although there seems to be no association between overall opium use and CRC, the risk of CRC might be increased among the persons who use opium more than twice a day.

Authors' contributions

MH did the literature review. HR, AN, MGH, MM, EM, RS, MS, BH, MB, RA, VA, SSH, AN, FN, AM, and AR contributed to data collection. FZ, RM, and MN provided clinical consultation. MH, HP, AR, RM, PB, EW, FK, KZ, and EP designed the study. MH and EP did the data analysis, interpreted data, and prepared the manuscript draft. All authors critically appraised the drafts of the manuscript and approved the final version. KZ is the guarantor of the study.

Authors' disclaimer

Where authors are identified as personnel of the International Agency for Research on Cancer/World Health Organization, the authors alone are responsible for the views expressed in this article, and they do not necessarily represent the decisions, policies, or views of the International Agency for Research on Cancer/World Health Organization.

Ethical approval

The study was approved by the Ethics Committee of the National Institute of Medical Research Development (NIMAD) in Iran (Code: IR.NIMAD.REC.1394.027).

Consent to participate

Written informed consent was obtained from the parents.

Disclosure statement

There is no competing interest between authors.

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Data availability statement

The data underlying this article cannot be shared publicly due to privacy of individuals that participated in the study. The data may be shared on reasonable request to the corresponding author.

References

[1] Sung H, Ferlay J, Siegel RL, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2021;71(3):209–249. doi: 10.3322/caac.21660.
[2] Xi Y, Xu P. Global colorectal cancer burden in 2020 and projections to 2040. Transl Oncol. 2021;14(10):101174. doi: 10.1016/j.tranon.2021.101174.
[3] Ferlay J, Colombet M, Soerjomataram I, et al. Cancer statistics for the year 2020: an overview. Int J Cancer. 2021;149(4):778–789. doi: 10.1002/ijc.33588.
[4] Power AM, Talley NJ, Ford AC. Association between constipation and colorectal cancer: systematic review and meta-analysis of observational studies. Am J Gastroenterol. 2013;108(6):894–903; quiz 904. doi: 10.1038/ajg.2013.52.
[5] Fosnes GS, Lydersen S, Farup PG. Drugs and constipation in elderly in nursing homes: what is the relation? Gastroenterol Res Pract. 2012;2012:290231–290237. doi: 10.1155/2012/290231.
[6] Roshandel G, Ghanbari-Motlagh A, Partovipour E, et al. Cancer incidence in Iran in 2014: results of the iranian national population-based cancer registry. Cancer Epidemiol. 2019;61:50–58. doi: 10.1016/j.canep.2019.05.009.
[7] World Drug Report. United Nations publication, Sales No. E.20.XI.6 [Internet]. 2020. Available from: https://www.unodc.org
[8] Rahimi-Movaghar A, Gholami J, Amato L, et al. Pharmacological therapies for management of opium withdrawal. Cochrane Database Syst Rev. 2018;2018(6):CD007522. doi: 10.1002/14651858.CD007522.pub2.
[9] IARC. Opium consumption. IARC Monogr Identif Carcinog Hazards Hum. 2021;126:1–253.
[10] Hayatbakhsh MM, Oghabian Z, Conlon E, et al. Lead poisoning among opium users in Iran: an emerging health hazard. Subst Abuse Treat Prev Policy. 2017;12(1):43. doi: 10.1186/s13011-017-0127-0.
[11] Beattie AD, Briggs JD, Canavan JSF, et al. Acute lead poisoning: five cases resulting from self-injection of lead and opium. QJM Int J Med. 1975;44:275–284.
[12] Masoudi M, Zali MR, Ehsani M, et al. Abdominal pain due to lead-contaminated opium: a new source of inorganic lead poisoning in Iran. Arch Iran Med. 2006;9:72–75.
[13] Sundbøll J, Thygesen SK, Veres K, et al. Risk of cancer in patients with constipation. Clin Epidemiol. 2019;11:299–310. doi: 10.2147/CLEP.S205957.
[14] Momen-Heravi F, Babic A, Tworoger SS, et al. Periodontal disease, tooth loss and colorectal cancer risk: results from the nurses' health study. Int J Cancer. 2017;140(3):646–652. doi: 10.1002/ijc.30486.
[15] IARC Monographs Vol 126 Group. Carcinogenicity of Opium Consumption. Lancet Oncol. 2020;21:1407–1408.
[16] Naghibzadeh-Tahami A, Feyzabadi VY, Khanjani N, et al. Can opium use contribute to a higher risk of colorectal cancers? A matched case-control study in Iran. Iran J Public Health. 2016;45:1322–1331.
[17] Khosravizadegan Z, Naghibzadeh-Tahami A, Akbari M, et al. Opium use and risk of lower gastrointestinal cancers: population-Based Case-Control study in South of Iran. Int J Cancer Manag. 2017;10(6):e8227. doi: 10.5812/ijcm.8227.
[18] Sheikh M, Shakeri R, Poustchi H, et al. Opium use and subsequent incidence of cancer: results from the golestan cohort study. Lancet Glob Health. 2020;8(5):e649–60–e660. doi: 10.1016/S2214-109X(20)30059-0.
[19] Hadji M, Rashidian H, Marzban M, et al. The iranian study of opium and cancer (IROPICAN): rationale, design, and initial findings. Arch Iran Med. 2021;24(3):167–176. doi: 10.34172/aim.2021.27.
[20] Hampel H, Frankel WL, Martin E, et al. Screening for the lynch syndrome (hereditary nonpolyposis colorectal cancer). N Engl J Med. 2005;352(18):1851–1860. doi: 10.1056/NEJMoa043146.
[21] Peltomäki P, Olkinuora A, Nieminen TT. Updates in the field of hereditary nonpolyposis colorectal cancer. Expert Rev Gastroenterol Hepatol. 2020;14(8):707–720. doi: 10.1080/17474124.2020.1782187.
[22] Kauppinen T, Toikkanen J, Pukkala E. From cross-tabulations to multipurpose exposure information systems: a new job-exposure matrix. Am J Ind Med. 1998;33(4):409–417. doi: 10.1002/(SICI)1097-0274(199804)33:4<409::AID-AJIM12>3.0.CO;2-2.
[23] Kauppinen T, Uuksulainen S, Saalo A, et al. Use of the finnish information system on occupational exposure (FINJEM) in epidemiologic, surveillance, and other applications. Ann Occup Hyg. 2014;58:380–396.
[24] Pukkala E, Guo J, Kyyrönen P, et al. National job-exposure matrix in analyses of census-based estimates of occupational cancer risk. Scand J Work Environ Health. 2005;31(2):97–107. doi: 10.5271/sjweh.856.
[25] Poustchi H, Eghtesad S, Kamangar F, et al. Prospective epidemiological research studies in Iran (the PERSIAN cohort study): rationale, objectives, and design. Am J Epidemiol. 2018;187(4):647–655. doi: 10.1093/aje/kwx314.
[26] Keshtkar AA, Semnani S, Pourshams A, et al. Pictogram use was validated for estimating individual's body mass index. J Clin Epidemiol. 2010;63(6):655–659. doi: 10.1016/j.jclinepi.2009.08.014.
[27] Etminan M, Collins GS, Mansournia MA. Using causal diagrams to improve the design and interpretation of medical research. Chest. 2020;158(1S):S21–S28. doi: 10.1016/j.chest.2020.03.011.
[28] VanderWeele T. Explanation in causal inference: methods for mediation and interaction. New York, NY: Oxford University Press; 2015.
[29] Mohebbi E, Rashidian H, Tahami AN, et al. Opium use reporting error in case-control studies: neighborhood controls versus hospital visitor controls. Med J Islam Repub Iran. 2021;35:60–67.
[30] Rashidian H, Hadji M, Marzban M, et al. Sensitivity of self-reported opioid use in case-control studies: healthy individuals versus hospitalized patients. PLoS One. 2017;12(8):e0183017. doi: 10.1371/journal.pone.0183017.
[31] Nemati A, Jafari S, Afshari M, et al. Comparing blood lead level among oral/inhaled opium addicts with a non-addict control group in the southeast of Iran. Addict Health. 2016;8:235–241.
[32] Wu Z, Han Y, Caporaso JG, et al. Cigarette smoking and opium use in relation to the oral microbiota in Iran. Microbiol Spectr. 2021;9(2):e00138-21. doi: 10.1128/Spectrum.00138-21.
[33] Karstensen JG, Burisch J, Pommergaard H-C, et al. Colorectal cancer in individuals with familial adenomatous polyposis, based on analysis of the danish polyposis registry. Clin Gastroenterol Hepatol. 2019;17(11):2294–2300.e1. doi: 10.1016/j.cgh.2019.02.008.
[34] Bodmer WF, Bailey CJ, Bodmer J, et al. Localization of the gene for familial adenomatous polyposis on chromosome 5. Nature. 1987;328(6131):614–616. doi: 10.1038/328614a0.
[35] Sohrabi M, Gholami A, Azar MH, et al. Trace element and heavy metal levels in colorectal cancer: comparison between cancerous

and non-cancerous tissues. Biol Trace Elem Res. 2018;183(1):1–8. doi: 10.1007/s12011-017-1099-7.

[36] Etemadi A, Hariri S, Hassanian-Moghaddam H, et al. Lead poisoning among asymptomatic individuals with a long-term history of opiate use in golestan cohort study. Int J Drug Policy. 2022;104: 103695. doi: 10.1016/j.drugpo.2022.103695.

[37] Botteri E, Iodice S, Raimondi S, et al. Cigarette smoking and adenomatous polyps: a meta-analysis. Gastroenterology. 2008;134(2): 388–395. doi: 10.1053/j.gastro.2007.11.007.

[38] Mohebbi E, Kamangar F, Rahimi-Movaghar A, et al. An exploratory study of units of reporting opium use in Iran: implications for epidemiologic studies. Arch Iran Med. 2019;22: 541–545.