RESEARCH ARTICLE

Risk Factors for Gallbladder Cancer in Nepal - a Case Control Study

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Abstract

Background: Gall bladder cancer (GBC) is highly fatal disease with poor prognosis, with a 5 year survival rate of <10%. It is relatively rare cancer worldwide; however it is the sixth cancer and second most common gastrointestinal tract cancer in Nepalese women. The study focused on associations of certain demographic, lifestyle, dietary, and reproductive factors with gall bladder cancer. Materials and Methods: We conducted a hospital-based matched case control study on newly diagnosed cases of primary GBC at BP Koirala Institute of Health Sciences and BP Koirala Memorial Cancer Hospital. Controls were healthy non-GBC relatives of cancer patients, matched for age, sex and marital status (in case of females) with cases at a ratio of 1:2. Data were collected between April 2012-April 2013 by semi structured interview from both cases and controls. Analyses were carried out with SPSS. Conditional logistic regression was used to find odds ratios and 95% confidence intervals for bivariate and multivariate analysis. Results: A total of 50 cases and 100 controls were enrolled in this study. On bivariate analysis, factors found to be significantly associated with gallbladder cancer were illiteracy (OR= 3.29, CI= 1.06-10.2), history of gallstone disease (OR=27.6, CI=6.57, 115.6), current smoker (OR=2.42, CI=1.005-5.86), early menarche <13 years (OR=2.64, CI=1.09-6.44), high parity more than 3 (OR=3.12, CI=1.25,7.72), and use of mustard oil (OR=3.63, CI=1.40, 9.40). A significant protective effect was seen with high consumption of fruits at least once a week (OR=0.101, CI=0.03-0.35). On multivariate analysis, history of gallstone disease, early menarche, current smoker and high consumption of fruits persisted as significant factors. Conclusions: History of gallstone disease, cigarette smoking and early menarche were associated with increased risk of gallbladder cancer while high consumption of fruits was found to have a protective effect.

Keywords: Gallbladder cancer - risk factors - gall stones - smoking - reproductive parameters - fruits - Nepal

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Introduction

Gallbladder cancer (GBC) is a rare cancer with incidence of <2 per 100,000 population worldwide (Stinton and Shaffer, 2012). Though the cancer is rare, it is fifth most common malignancy of gastrointestinal tract in United States of America (USA) and most common malignant tumor of biliary tract, representing 46 % of all such malignancies. It is highly fatal disease with poor prognosis. The diagnosis is commonly made at an advanced stage because of the vague signs and symptoms of disease. The overall mean survival rate for patients with advanced gallbladder cancer is 6 months, with a 5-year survival rate of 5% (Stinton and Shaffer, 2012).It is predominantly a disease of elderly women with peak incidence in the seventh decade of life. The reported age range at presentation is 40-80 years with men: women ratio ranging from 1:2 to 1:3.2 (Pandey, 2003).

The disease has shown marked geographical variation, being low in several European countries and the USA, relatively high in selected central European countries, and very high in some countries of Latin America and Asia. Chile (Valdivia) was found to have the highest incidence of gall bladder cancer, 12.3 per 100,000 among and 27.3 per 100,000 populations among men and women respectively (ASW, age-standardized to the world population) (Eslick, 2010). It also shows marked variation in ethnicity, higher incidence rates are seen among Chileans Mapuche Indians Hispanics, Bolivians, North American Indians, Mexican-Americans, and Central Europeans (Lazcano et al., 2001).

In context of Nepal, it is the sixth most common cancer in females after cervix, breast, lung, ovary, and stomach (Pun et al., 2015). In a study by Shrestha et al on cases with cholecystectomy, incidence of gall bladder carcinoma was found to be 3.3% out of which 1.4% were incidental finding and most of them were associated with gall stone (80%) (Shrestha et al., 2010).

The exact etiology of the gallbladder carcinoma is

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obscure, with more than one factor being postulated for the pathogenesis of this cancer. A strong association of gallstone disease and gallbladder cancer has been observed, suggesting it to be the important risk factor for pathogenesis of cancer. Similarly, increasing age and the female gender are also important risk factors. In addition, smoking, obesity, parity, genetic, chronic bacterial infection, low socioeconomic status, dietary habit, and benign neoplasm of the gallbladder are also postulated as risk factors for pathogenesis of gallbladder cancer. (Dwivedi et al., 2013; Khan et al., 2013; Panda et al., 2013; Wenbin et al., 2013; Nogueira et al., 2015; Iyer et al., 2016)

Though gallbladder cancer is relatively common in Nepal, not much work has been done to find the risk factors associated with it. It could be due to prolonged exposure with gallstone as gallstone disease is prevalent in younger age or it could be associated with other lifestyle and reproductive factors like smoking, alcohol, number of children, menarche etc.

Thus, this study tried to elicit the associated factors in the development of the gallbladder cancer in Nepal.

Materials and Methods

A hospital based matched case control study was carried out at BP Koirala Memorial Cancer Hospital (BPKMCH) and BP Koirala Institute of Health Sciences (BPKIHS) from April 2012- April 2013. BPKMCH is the first national cancer hospital which receives cancer patients referred from 75 districts of the country and provides care to more than 10000 people annually. BPKIHS is a tertiary care university hospital which serves patients referred from Eastern and Central Region of the country. Cases were patients newly diagnosed with gallbladder carcinoma by histological or cytological examination. Controls were healthy people without any history of malignancy, accompanying the patient having cancer other than gallbladder, matched with cases by age (±2), sex and marital status (in case of female) in ratio of 2:1. The research protocol was approved by the Institutional Ethical Review Board of BPKIHS. Informed consent was taken from each participant before collection of data.

Data were collected using the pretested semi structured questionnaire regarding socio demographic, lifestyle factors, reproductive factors, and dietary factors. A semi quantitative food frequency questionnaire was used to measure the role of dietary factors. Weight and height were determined by anthropometric measurements. Variables included in lifestyle factors were history of tobacco, smoking and alcohol consumption with frequency, type, and duration for these three factors.

Participants who had consumed alcohol at least once a week for at least 6 months were considered as ever drinker otherwise as never drinker. The amount of alcohol consumed was converted in grams per week (Babor et al., 2001; Pradhan et al., 2012). In case of smoking, participants who had smoked cigarettes, cigars or pipe regularly at the time of interview or stopped within one year of the interview were considered as current smokers. Participants who used to smoke cigarettes, cigars or a pipe regularly but quitted at least one year prior to interview were considered as past smokers. Participants who never smoked cigarettes, cigars, pipe, any other tobacco product, or reported a smoking frequency of less than 100 times over a life time were considered never smoker. BMI was calculated as weight in kg/ height in meter square. They were categorized as underweight (<18.5), normal (18.5-24.9), and overweight (≥25).

A detailed dietary history was obtained from each participant. We enquired history of usual diet, 2 year preceding the onset of symptoms to rule out possible changes in diet because of symptoms and treatment, assuming that this reflects the lifetime dietary habit of the study participants.

The commonly consumed food items included were cereals (wheat/rice), pulses/ legumes, vegetables, fruits, milk, milk products, animal foods (red meat/fish/ poultry), tea, cooking medium etc.

In case of female, reproductive history regarding age at menarche, age at menopause, parity, number of miscarriage, age at first pregnancy, age at first child birth, age at last child birth, and methods of contraceptive used were obtained.

Sample size calculation was done with nMaster 2.0. From previous study, smoking was found to be important risk factor for gallbladder cancer, the odd ratio (OR) among smokers was 2.71 (Khan et al., 1999) and prevalence of smoking was 32% (World Health Organization, 2010).

Table 1. Sociodemographic Characteristics of Gallbladder Cases and Control

Characteristics	Cases (N=50) %	Controls (N=100) %	P value
Sex			1
Male	28	28	
Female	72	72	
Age categories			
Less than 40	12	11	
40-49	14	16	
50-59	38	37	0.99
60-69	24	25	
More than 70	12	11	
Mean±SD	54.5±11.0	54.3±10.6	
Hospital			
BPKMCH	84	76	0.26
BPKIHS	16	24	
Religion			
Hindu	88	85	0.61
Others	12	15	
Occupation			
Housemaker	56	42	0.26
Agriculture worker	24	35	
Business	4	9	
Service	4	8	
Skilled worker	6	4	
Others	6	2	
Per Capita Income			
Below Poverty Line	60	70	0.279
Above Poverty Line	40	30	

Assuming 95% Confidence level, 80% power and ratio between case and control as 1:2, the estimated sample size for matched study was 50 cases and 100 controls with total of 150 participants.

Data were entered in Microsoft excel sheet and analyzed in SPSS17.0. Conditional logistic regression was used to calculate the matched odds ratio. The independent variables with p value <0.20, were considered for the multivariate conditional logistic regression analysis. Forward selection method was used to determine the variable for model. The final model included education, type of hospital, history of gallstones disease, smoking status, fruits consumption, place of residence etc. A separate analysis was done for females, which included reproductive factors, history of gallstone disease, and education. A p-value of <0.05 was considered as statistically significant.

Results

All together 150 sample with 50 cases and 100 controls in the ratio of 1:2 were included in the study. Out of all cases, 84% were interviewed in BP Koirala Memorial Cancer Hospital (BPKMCH) and 16% in BP Koirala Institute of Health Sciences (BPKIHS) where as 76% and 24% of the controls were interviewed in BPKMCH and BPKIHS respectively.

About two third (72%) of cases were female with male: female ratio of 1:2.57 and all of them were married. The mean age of the gallbladder cancer patient was 54.4 ± 11.02 years. Most of them were of age group 50-59 years followed by age group 60-69 years. Most of the participants were Hindu (88% cases and 85% controls) by religion. By occupation both groups were comparable, majority of them were house makers and agriculture workers. In our study, 84% of cases and 70 % of controls were from terai region and remaining 16% and 30% from the hill/mountain regions respectively (table 1).

Eighty six percent of cases were illiterate whereas 72% of controls were illiterate. There was significant increased odds of cancer among illiterate people than in literate (OR= 3.29, CI= 1.06-10.19).

Majority of participants with gallbladder cancer had gallstone disease (70%) than controls (14%). People with history of gallstone had significant increased risk (OR=27.6, CI=6.57-115.6) of gallbladder cancer. The

Table 2. Table Showing Lifestyle, Diet and Medical History in Relation With Gallbladder Cancer

Characteristics	Cases (N=50) %	Controls (N=100) %	OR	95% CI	P value	Adjusted OR#	95% CI	P value
Education								
Literate	14	28	1	reference	0.04			
Illiterate	86	72	3.29	(1.06,10.2)				
Smoking Status								
Never	56	66	1	reference		1	reference	
Former	16	21	1.001	(0.38,2.62)	0.99	0.8	(0.14, 4.42)	0.80
Current	28	13	2.42	(1.005,5.86)	0.049	5.28	(1.06,26.3)	0.04
Pack Year								
None	56	66	1	reference				
0.1-20	30	29	1.25	(0.53,2.90)	0.6			
20.1+	14	5	2.99	(0.92,9.72)	0.06			
Smokeless Tobacco								
No	76	78	1	reference	0.76			
Yes	24	22	1.15	(0.46,2.86)				
Alcohol								
NO	70	75	1	reference	0.53			
YES	30	25	1.26	(0.61,2.59)				
Amount in Grams								
None	70	75	1	reference				
1-19.9	10	11	0.92	(0.28,2.97)	0.88			
20+	20	14	1.5	(0.62,3.63)	0.36			
Gall Stone								
absent	30	86	1	reference	<0.001	1	reference	<0.001
present	70	14	27.6	(6.57,115.6)		46.4	(6.92,311.3)	
Enteric Fever in Past								
No	14	10	1	reference				
Yes	86	40	1.4	(0.53,3.67)	0.49			
Cooking Medium								
Refined oil(R)	12	35	1					
Mustard oil(M)	68	53	3.63	(1.40,9.40)	<0.01			
M+R	20	12	4.59	(1.38,15.2)	0.01			
Dietry Pattern								
Vegetarian	20	18	1		0.14			
Non vegetarian	80	82	2.41	(0.75,7.73)				
Fruits								
<a day/week	38	10	1	reference		1	reference	0.02
At least one day/week	62	90	0.101	(0.03,0.35)	<0.001	0.07	(0.09,0.70)	

Table 3. Relationship of Menstrual and Reproductive Factors and Gallbladder Cancer

Characteristics	Cases (N=36) %	Controls (N=72) %	OR	95% CI	P value	Adjusted OR#	95% CI	P value
Age at Menarche								
>13	55.6	76.4	1	reference	0.03	1	reference	0.02
≤13	44.4	23.6	2.64	(1.09,6.44)		14.8	(1.41,154.3)	
Age at First Pregnancy								
>20	19.4	37.5	1	reference	0.06	1	reference	0.89
≤20	80.6	62.5	2.46	(0.93,6.48)		1.18	(0.18,6.86)	
Age at Last Child								
≤30	33.3	50	1	reference	0.07	1	reference	0.08
>30	66.6	50	2.4	(0.93,6.21)		6.09	(0.82,45.3)	
No of Children								
≤3	19.4	44.4	1	reference	0.01	1	reference	0.6
>3	80.6	55.6	3.11	(1.25,7.72)		1.47	(0.33-6.48)	
No of Miscarriage								
≤1	80.6	88.9	1	reference	0.24			
>1	19.4	11.1	1.93	(0.63,5.91)				
No. of Time Pregnant								
≤3	33.3	13.9	1	reference	0.04			
>3	66.7	86.1	2..95	(1.04,8.36)				
	N= 30	N=54						
Age at Menopause								
≤46	43.3	51.9	1	reference	0.14			
>46	56.7	48.1	2.17	(0.77,6.08)				
Duration of Menstruation								
Mean± SD	33.0±4.04	31.0±5.12						

adjusted for history of gallstones, education, and each other

radiographic report was available for 28 patient out of 32 cases and the average size of largest stone documented was 15.13 mm (SD=5.77) with range 4.3-35 mm. In the radiographic report, there was no consistency in reporting the actual number of the stone, so the data was recorded as single stone or multiple stones and majority of them had multiple stone (75%). On observing the reports, only one cancer case had gallbladder pathology that was porcelain gallbladder.

About half (44%) of the cases and 34% of the controls were smokers. In comparison to non smokers, current smokers had 2.4 times significant risk of GBC (OR=2.42, CI=1.005-5.86). On an average the cases started smoking 5 years earlier than the controls. Likewise, the exposure to smoking was also longer in cancer patients. The cancer patients smoked tobacco for an average of 33 years and controls smoked for an average of 26.7 years, though it was not significant. Dose dependent risk for GBC was observed with number of cigarette and pack year of cigarette smoking as compared with non-smokers. No significant risk was observed with tobacco chewing and alcohol drinking habits.

The BMI of the control group (mean± SD= 24.5±4.84) was higher than the case group (mean± SD= 19.2±4.36).

Early age at menarche (age < 13 years, OR= 2.64, CI=1.09-6.44), higher number of pregnancy (gravid > 3 OR= 2.95, CI=1.04-8.36), and higher number of children (parity > 3, OR=3.11, CI=1.25-7.72) were found to have significantly higher risk of gallbladder cancer. Similarly, early age at pregnancy, late age at last child birth, and late age at menopause also increased risk of gallbladder cancer but was not found to be statistically significant (table 3).

In comparison with other refined oil users, significant elevated risk was observed (OR=4.59, CI=138.-15.14) among mustard oil users as a cooking medium. High fruit consumption showed protective effect. In comparison with fruits consumption less than a day/week, high consumption (at least once a weak) of fruits had significant protective effect (OR=0.101, CI=0.03-0.34)

In a multivariate analysis history of gallstone disease (present vs. absent) (OR=46.4, CI= 6.92-311.3), current smokers (OR= 5.82. CI=1.06-26.3) and early age at menarche in female (<13 and ≥13) (OR= 14.8, CI=1.41-154.3) were found to be independent risk factors. Similarly, consumption of fruits (at least once a week as compared to less than once a week (OR=0.07, CI= 0.09-0.70) was found to be independent protective factor for the gallbladder cancer.

Discussion

In the present study, mean age of the study participants in case group was 54.4 years with 72% female. The finding is similar to the retrospective analysis of 52 patients in BPKMCH by Jianzhong Wang in which the mean age of gallbladder cancer patient was 51.5 years and more than 80 percent of cases were female (Wang et al., 2006). Similar finding had been reported in several studies from Indian subcontinent which showed gallbladder cancer affects predominantly female in their fourth and fifth decade of life. (Pandey, 2003; Kumar et al., 2006; Alvi et al., 2011; Panda et al., 2013) However, studies from other part of the world has shown that gallbladder cancer was predominant in female with peak incidence in sixth and seventh decade of life (Arminski, 1949; Nervi et al., 1988; Kato et al., 1989; Zatonski et al., 1997; Scott et al., 1999; Lazcano-Ponce et al., 2001). The reason, for gallbladder cancer developing at a younger

age in Indian subcontinent population including Nepal, could be that gall stones develops at a younger age in Indian subcontinent population than western population (Kapoor and McMichael, 2003; Shrestha et al., 2010; Alvi et al., 2011) and gallstone is important risk factor for development of cancer. A study by Shrestha et al on cholecystectomy patients reported that gallstone is most common in 3rd decade of life (Shrestha et al., 2010). This age difference of gallbladder cancer patients from western population might be over-estimated, as the life expectancy in Nepal is low (male 68 and female 70 years) (World bank, 2011).

The sex ratio (F/M) of incidence rates varied greatly worldwide. In majority of countries, ratio was between 2 and 3. In several high-risk areas (e.g. Pakistan, India, Colombia and Spain) as well as in few selected low-risk areas (e.g. Denmark) ratio was > 5 and in countries like Korea, Japan and some parts of China it was close to one (Randi et al., 2006). Similar to other majority of the countries, F/M ratio was 2.57 in the current study. A history of gallstones had been established as most important risk for gallbladder cancer. A wide range of association had been reported in several case control studies from OR= 2.3 (95% CI: 1.2-4.4) to OR=34.4 (95% CI: 4.5-266) (Strom et al., 1995; Zatonski et al., 1997; Khan et al., 1999; Hsing et al., 2007; Tyagi et al., 2008; Alvi et al., 2011). This study also confirmed gallstone as the strong independent risk factor of gallbladder cancer. Similarly, this association had also been observed in several cohort studies, however a Japanese cohort study by Yagyu et al found no association between cancer and gallstone (Randi et al., 2006). Presence of gallstone causes repeated trauma to the gallbladder mucosa leading to a state of chronic inflammation. This Chronic inflammation leads to metaplasia which will progress from stage of dysplasia, carcinoma in situ and eventually invasive adenocarcinoma (Barreto et al., 2014; Li et al., 2014).

Increasing size or more numbers of stone had been shown to be associated with an increased risk of cancer. Stone size > 3cm had been shown to carry risk of 9 to 10 times to develop gallbladder cancer as compared to size less than 1 cm. (Csendes et al., 2000; Shrikhande et al., 2010) In agreement with literature, large size of stone, with an average size of 15.13mm, and multiple stone were associated with GBC patients in current study.

The relationship of smoking and gallbladder cancer has been mixed. Some of the studies suggested increased risk (Khan et al., 1999; Shukla et al., 2008; Tyagi et al., 2008; Yagyu et al., 2008), while other suggested no association of smoking in gallbladder cancer. (Zatonski et al., 1997; Pandey and Shukla, 2003).There was only one prospective report evaluating the effect of smoking on GBC, which found that female current smokers had increased risks of death because of GBC, whereas male current smokers and ex-smokers had a similar risk of GBC. (Yagyu et al., 2008) A recent meta-analysis has reported that smokers had a 45% increased risk of developing GBC, which was independent of alcohol use and a history of gallstones (Wenbin et al., 2013). This study also showed that the risk of gallbladder cancer increased among the current smokers independent of history of gall stones and less fruit consumption. The cases started smoking around five year earlier and smoked for longer duration than the controls. The excess risk of cancer was also observed with increasing frequency and pack year of smoking. This suggests that smoking has role in initiation of carcinogenesis as well in progression of the disease. After being absorbed from the lungs, tobacco smoke compounds such as nitrosodimethylamine, formaldehyde, benzene, nicotine and chromium which are thought to reach the liver or pancreas through the blood flow. In the same way, deleterious compounds of tobacco smoke carried through the blood flow may accumulate in bile, exerting carcinogenic effects on the gallbladder epithelial cells.

In contrast to previous finding, we observed inverse relation between BMI and gallbladder cancer. (Strom et al., 1995; Zatonski et al., 1997; Larsson and Wolk, 2007). This contradiction could be the result of difference in measurement method. We measured weight and height at the time of interview, the weight may have been an effect of cancer malnutrition (Rai et al., 2006) rather than the cause of gallbladder cancer. Similar finding was also reported in a study from Delhi, which also measured weight at the time of interview and found inverse relationship with weight (Tyagi et al., 2008).

Parity has appeared as a risk factor for gallbladder cancer in many literatures. High parity and higher number of pregnancy had been related to gallbladder cancer risk in several previous studies. The odds ratio ranged between 1.3 and 9.02, and between 1.0 and 6.7, respectively depending on the level of parity and gravida considered. (Moerman et al., 1994; Zatonski et al., 1997; Pandey and Shukla, 2003; Rizvi and Zuberi, 2003; Randi et al., 2006; Shukla et al., 2008; Andreotti et al., 2010; Panda et al., 2013). In the line of previous studies, current study also showed the risk of gallbladder cancer with high parity and higher number of pregnancy. It is proposed that the elevated level of estrogen and progesterone hormones during pregnancy alter the lithogencity of bile, predisposing to gallstone formation and probably carcinoma of the gallbladder. Gall bladder stasis during pregnancy may also promote the formation of stone and sludge. (Moerman et al., 1994)

Age at menarche has been controversial as the risk factor for gallbladder cancer. Some authors have shown late age (Pandey and Shukla, 2003; Andreotti et al., 2010) and others have shown early age at menarche to be risk factor (Moerman et al., 1994; Shukla et al., 2008; Jain et al., 2013; Panda et al., 2013). In the present study, early age at menarche was found to be independent risk factor for gallbladder cancer. Likewise, cases, who had already attained menopause, had menopause at older age and longer duration of fertility (i.e. menstruation cycle) than controls. These all point to longer active ovarian functioning and thus longer exposure to female sex hormones. Human gallbladder contains both estrogen and progesterone receptors, and higher expression of these receptors and its variant has been reported in gallbladder cancer patient suggesting important role of sex hormones in gallbladder cancer susceptibility. (Gupta et al., 2012; Srivastava et al., 2012; Sharma et al., 2014)

Among the dietary factors, high intake of fruits was

found to be protective factor for gallbladder cancer. Similar results have been reported in previous studies. (Kato et al., 1989; Pandey and Shukla, 2002; Rai et al., 2005) The protective effect of fruits against cancer appears to depend on multiple constituent substances and mechanisms. A large number of potentially anti-carcinogenic agents are found in fruits, including carotenoids, vitamins C, vitamin E, selenium, folic acid, dietary fiber, dithiolthiones, isothiocyanates, glucosinolates, indoles, phenols, flavinoids, protease inhibitors, allium compounds, plant sterols and limonene. These have complementary and overlapping mechanisms of action, which include the induction of detoxification enzymes, inhibition of nitrosoamine formation, dilution and binding of carcinogens in the digestive tract, alteration of hormone metabolism and antioxidant effects (Steinmetz and Potter, 1991).

Unlike the study by Pandey et al which showed no association with mustard oil and gallbladder cancer (Pandey and Shukla, 2002), the significant risk was observed with use of mustard oil as cooking medium for gallbladder cancer in our study. The risk was observed among those who were using mustard oil regularly or sometimes. Similarly, a cross-sectional study in India has shown more than 90 percent of the cases used the mustard oil regularly (Dwivedi et al., 2013). Several animal studies suggests that mustard oil adulterants, argemone oil and butter yellow, could be one of the etiological factors in the development of gall bladder adenocarcinoma and adenoma, and that argemone oil induced gallbladder cancer through gallstone formation, while butter yellow induced gallbladder cancer is independent of stone formation (Mishra et al., 2012). Similarly, another study by Dixit et al demonstrated an increased concentration of mustard oil adulterants like sanguinarine and diethylnitrosamine, in blood and tissue of gallbladder cancer patients compared to patients with cholelithiasis (Dixit et al., 2013). It suggests that the association of mustard oil with GBC is probably due to its adulteration. There was also a report of adulteration of mustard oil with argemone oil and outbreak of dropsy from bordering state of India. (Sharma et al., 1999)

There are few potential limitations in present study. Recall bias may have effect on associations, especially for dietary and lifestyle factors, as cases may have better recall than controls. As the interviewer was not blinded, interviewer bias cannot be totally excluded. Another limitation might be selection bias as our study was hospital-based in a major hospitals, which are also tertiary referral centers. Our control series was not population-based, thus it is possible that the distribution of the factors of interest do not reflect the distribution in the base population. Hospital-based controls from the surrounding hospitals were difficult to match with geographic region since people from various regions of Nepal come to the cancer hospital for treatment.

We have attempted to identify some of the risk factors for gallbladder cancer through this case control study. The study indicates selected lifestyle and reproductive factors play important role in development of gallbladder cancer in Nepal. The study found history of gallstone, cigarette smoking and early menarche associated with increased risk of gallbladder cancer. High consumption of fruits is found to be important protective factors. Further prospective study should focus on these aspects.

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