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Betel Quid Chewing: A Possible Source of Heavy Metal Exposure to the People in Khulna, Bangladesh

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Abstract

Betel quid (BQ) chewing is an ancient tradition across the globe. The present study aims to assess the non-carcinogenic and carcinogenic health risk of BQ chewers by estimating the concentration of heavy metals (Cr, Mn, Cu, Pb, Zn, Cd, and As) in *Piper betel* leaf (PBL) and Areca nut (AN). The concentrations of heavy metals were determined by atomic absorption spectrophotometry (AAS) technique and the health risks were calculated based on USEPA deterministic model. The average concentration of Mn, Cu, Zn, As, Cr, Cd, and Pb are observed 1.110, 3.250, 7.971, 0.561, 0.333, 0.084 and 0.323 mg kg^{-1} in PBL; 1.756, 5.147, 5.022, 0.081, 0.434, 0.094 and 0.224 mg kg^{-1} in AN respectively. Evaluation of the non-carcinogenic parameters (target health quotient and health index) for heavy metals never exceeded the USEPA risk limit (1.0), indicating no non-carcinogenic health effects. Likewise, the value of the carcinogenic parameter (Target cancer risk) was lower than the USEPA tolerable limit (10^{-4}) for PBL and AN, but ingestion of BQ (PBL+AN) poses slight concern for Cd-induced cancer risk. The overall analysis reveals that BQ chewing is almost safe for BQ chewers in Khulna, Bangladesh region, but Cd should be paid more attention as a risk source. Therefore, regular monitoring of heavy metals in different parts of Bangladesh should be done and necessary steps should be taken to control heavy metal pollution.

1. Introduction

The *Piper betel* leaf (PBL), a deep green heart-shaped leaf, is the oldest common masticatory used as a wrapper for the chewing of areca nut, or tobacco along with other ingredients where it is mainly named as betel quid (BQ, *Paan*) [1]. Areca nut (AN, *Supari*) is the seed of the oriental palm tree (*Areca catechu*). The major constituents of AN are fats, proteins, carbohydrates, polyphenols, mineral, crude fiber, alkaloids and [2]. Since primitive time, it is widely consumed by all ages groups in many parts of the world, especially in South and Southeast Asia [3]. BQ ranks as the fourth most consumed drug after tobacco, alcohol, and caffeine for its mild cholinergic and psychoactive effects [4]. Many BQ products in different parts of the world are not chewed; rather, they are placed in the mouth or applied to the oral cavity and remain in contact with the oral mucosa [2]. In Bangladesh, BQ is traditionally chewed not only as a habit but also as an item of rituals, etiquette, and manners. On formal occasions offering *Paan* symbolized the time for departure. It is also used in Hindu puja, wedding festivals and to visit relatives. Adult Bangladeshi women get together with *paandani* [1] along with friends and sing traditional Bangla folk songs "*Jodi sundor akta mukh paitam, sodorghater paan khili tare banai khauyaitam*", "*Paan khaiya thot lal korilam bondhu vaggo hoilo na*". Scientific research on the PBQ reveals that it possesses many beneficial bioactivities and its extract has a great potential to be used in developing commercial products. Several literatures reported that PBL contains a wide variety of biologically active compounds, vitamins, essential amino acids and have medicinal value [5, 6]. BQ chewing produces aromatic volatile oil, which has powerful antiseptic properties. Pharmacological effects of BQ chewing include the abundant flow of saliva; temporary dulled of taste perception, stimulation of muscular and mental efficiency, as well as diminished hunger and improved digestion [7]. In spite of many beneficial bioactivities, some literature reported BQ is not only an addictive substance and psycho-stimulant [8] but also a carcinogen [2, 9]. The World Health Organization (WHO) and the International Agency for Research on Cancer reported that chewing BQ may contribute to carcinogenic health risk to the human [2, 9, 10]. The main carcinogenic factor is believed to be areca nut. A recent study found that PBL-AN with and without tobacco increased oral cancer risk by 9.9 and 8.4 times, respectively [11]. In addition, epidemiological investigations have revealed that prolonged BQ use confers an increased risk of esophagus [12], hepatocellular carcinoma [13], liver cirrhosis [14], metabolic syndromes [15], diabetes mellitus [16], high blood pressure [17], cardiovascular disease [18], heart disease [19], chronic kidney disease [20], and adverse effects on mortality from cancer and from all causes [18].

Figure 1: (a) Cultivation of PBL (b) *Areca catechu* plant (c) Paandani (d-f) BQ stores

In several districts (Satkhira, Bagerhat) of Bangladesh, PBL cultivation is a common source of income for farmers (Figure 1). Due to geographical location, rapid industrialization, improper agricultural practice, vehicle emission, excessive use of wastewater, fertilizer and pesticides the growing soil might be polluted [21-25]. As a consequence, the cultivated PBL might also be contaminated with different types of toxic substances. Heavy metals are among the major contaminants and may consider the most important problem in our environment. Such a problem is getting more serious all over the world especially in developing countries like Bangladesh [21]. Generally, heavy metals are found in the earth crust, have long biological half-lives and they can persist for a long time in the environment without any biodegradation [22, 26, 27]. When the betel vine is cultivated, these metals might be deposited on the surface of roots, transport them upwards to their shoots and finally accumulate them inside the plant tissues [28]. Long-term consumption of BQ might arise the probability of the accumulation of heavy metals in the different organs [29] which might lead to unexpected side effects like cancer, kidney damage, bone diseases, cardiovascular diseases, etc [30, 31]. Moreover, proteins in the human body are easily attacked by heavy metals and lose their normal enzymatic activities. Heavy metals not only damage the human body but also affect the physiological functions of plants like chlorosis, growth of the plants, nutrient uptake, and reduction of nitrogen fixation and disorders of plant metabolism [32]. Several groups of researcher investigated the proximate compositions and vitamins content of PBL and AN [2, 7, 8, 33, 34]. To date, there are only a few reports published about the concentration of heavy metals in PBQ and AN in Bangladesh. As heavy metals pose negative impact on the human health, therefore, this study aims to determine the concentration of heavy metals (Cr, Mn, Cu, Pb, Zn, Cd, and As) in PBL and AN; to assess the associated health risk (carcinogenic and non-carcinogenic) of BQ chewers based on United States Environment Protection Agency (USEPA) deterministic approaches.

2. Materials and Methods

Generally, BQ is sold in the tea stall, varieties store, grocery shop, vegetable shop and roadside markets where the local people get together and make gossip. In this study, the BQ (n=300) samples were randomly collected from thirty individual stores (n=10) located in Khulna, Bangladesh. **Table 1** represents a general description of the sampling station.

Table 1: General description of the sampling station

Sampling Station	Sampling Location	Latitude	Longitude
S-01	Fulbari Gate Bazar	22°53'51.3"N	89°30'38.2"E
S-02	Relligate Stand	22°53'00.5"N	89°31'00.1"E
S-03	Daulatpur Bazar	22°52'13.5"N	89°31'32.6"E
S-04	Mohsin Mor Bus Stop	22°52'24.3"N	89°31'32.6"E
S-05	B.L. College More	22°51'57.5"N	89°31'24.4"E
S-06	Notun Rastha More	22°51'44.9"N	89°31'44.0"E
S-07	Goalkhali Bus stand	22°51'18.9"N	89°31'43.9"E
S-08	Navy School & College More	22°51'06.4"N	89°31'43.5"E
S-09	Khalishpur New Market Road	22°51'31.0"N	89°32'48.7"E
S-10	Alamnagar Bazar	22°51'01.8"N	89°32'43.8"E
S-11	Kadomtala Jame Masjid More	22°50'43.4"N	89°32'26.3"E
S-12	Boikali Bazar	22°50'29.9"N	89°32'15.0"E
S-13	Boyra Bazar	22°50'07.5"N	89°31'56.5"E
S-14	Khulna Medical College Hospital more	22°49'39.4"N	89°32'13.2"E
S-15	Sonadanga Bus Terminal	22°48'57.9"N	89°32'30.7"E
S-16	Khulna Paikary Kacha Bazar	22°48'41.5"N	89°32'26.1"E
S-17	Gollamari Bazar	22°48'01.1"N	89°32'23.2"E
S-18	Khulna Zero Point More	22°47'53.6"N	89°31'43.3"E
S-19	Dolkhola Bazar	22°48'19.5"N	89°33'37.2"E
S-20	Banorgati Kacha Bazar	22°48'27.1"N	89°32'51.4"E
S-21	KCC Shondha Bazar	22°48'43.8"N	89°33'24.8"E
S-22	7 Rasta More	22°48'37.1"N	89°33'39.5"E
S-23	Royal More	22°48'36.9"N	89°33'44.6"E
S-24	Shantidham More	22°48'44.4"N	89°33'40.4"E
S-25	Dak Bangla More	22°48'59.1"N	89°33'45.4"E
S-26	Railway station More	22°49'12.9"N	89°33'37.6"E
S-27	Shibbari Circle	22°49'20.1"N	89°33'10.7"E
S-28	Rupsha Shondha Bazar	22°48'06.0"N	89°34'47.0"E
S-29	KCC Women's College More	22°48'10.8"N	89°34'30.6"E
S-30	Shahid Hadis Park More	22°48'58.7"N	89°33'57.3"E

The PBL and AN were separated from the BQ samples and dried in an oven at 100±1°C until the constant weight is obtained. Then the samples were ground into finely powdered form. 1 g of each dry sample (PBL and AN) was digested by adopting wet digestion method described by Allen et al., [35] and the concentration of heavy metals (Cr, Mn, Cu, Zn, Pb, As and Cd) in individual digested solution was quantified by using an atomic absorption spectrophotometer (Shimadzu AA-7000, Tokyo, Japan) [21-23, 36].

The means, range of the concentrations of the heavy metals were calculated by using the Microsoft Excel 2010 and Statistical Package for Social Sciences (SPSS) version 16.0. Pearson's correlation was analyzed to establish specific relationships among the different metals as well as to distinguish the possible common sources of contamination and how some of the heavy metals influence their concentration.

2.1 Estimated Daily Intake (EDI)

Daily intake of contaminated vegetables is a general pathway of heavy metal exposure to the human. EDI of heavy metals from these foods can be calculated by using the equation [21, 37]:

$$EDI = \frac{H_{mc} \times R_i}{B_w} \quad (1)$$

Where, $R_i = W_{BQ} \times DI_{BQ}$; In this work, a survey was conducted among the 300 BQ chewers in Khulna, Bangladesh to estimate the daily intake of BQ. R_i denotes the rate of ingestion (0.016687 kg day^{-1}); W_{BQ} is the average weight of one BQ (0.002 kg dry weight), DI_{BQ} represents the average daily intake of BQ (8.33 BQ per day; H_{mc} is the concentration of heavy metal (mg kg^{-1} dry weight); B_w is the average body weight of Bangladeshi people (49.5 kg) [21, 38].

2.2 Target Hazard Quotient (THQ) and Health Index (HI)

The non-carcinogenic risks of BQ chewers were determined using the target hazard quotient (THQ). This is the ratio between the estimated daily intake (EDI) and reference dose (D_f) of a contaminant. It was described by using the following formula [26, 37]:

$$\text{THQ} = \frac{EDI \times E_f \times D_e}{D_f \times T_{avncar}} \quad (2)$$

Where, E_f is the exposure frequency (365 days $year^{-1}$); D_e denotes exposure duration (71.8 years, but in the study we have considered it 51.8 years. Generally, Bangladeshi people start BQ chewing at the age of 20 years); Reference dose (D_f) of Cr, Mn, Cu, Zn, Pb, Cd and As are 1.5, 0.14, 0.04, 0.30, 0.0035, 0.003 and 0.0003 (mg $kg^{-1}day^{-1}$) respectively [39-41]; and T_{avncar} implies average time for non-carcinogens (365 days $year^{-1} \times D_e$) [40]. Hazard Index is the sum of Hazard quotients of all metals. It was calculated by the formula [13, 42].

$$\text{HI} = \sum \text{THQ} = \text{THQ}_{(Mn)} + \text{THQ}_{(Cr)} + \text{THQ}_{(Cu)} + \text{THQ}_{(Zn)} + \text{THQ}_{(Pb)} + \text{THQ}_{(Cd)} + \text{THQ}_{(As)} \quad (3)$$

2.3 Target Cancer Risk (TCR)

TCR was estimated by using the equation [37]:

$$\text{TCR} = \text{THQ} \times S_{cpo} \quad (4)$$

The reference values of carcinogenic potency slope (S_{cpo}) of Pb, Cd and As are 0.0085, 6.1 and 1.5 mg kg^{-1} body weight $days^{-1}$ respectively [39].

3. Results and Discussion

BQ chewing is a prehistoric tradition of the Bangladeshi population. If PBQ and AN contain traces of heavy metals, the BQ chewers might be susceptible towards carcinogenic and non-carcinogenic health hazards. When the betel vine is cultivated in a polluted environment, the plant might absorb heavy metals from the air, water, and soil which might pose adverse effects on environmental health. Heavy metal pollution is a menace to our environment as they are foremost contaminating agents of our food chain [43]. Depending on the chemical nature, heavy metals are classified as toxic and essential elements. The potentially toxic metals such as Cd, Pb, As, and Cr might exert toxic and adverse health effects even at low concentration [44, 45] therefore entry of these metals in the food chain is a major concern. On the other hand, essential elements are those which are needed to be consumed in an adequate amount to maintain the normal physiological functions of human [44, 46]. Some heavy metals like Cu, Zn, and Mn play a significant role in plant physiology. But if metal intake exceeds the permissible limit or ingests for a long time, these elements may pose deleterious effects to the human [47]. To assess the health risks of the BQ chewers in Khulna, Bangladesh, the concentration of heavy metals in PBL and AN have been estimated and displayed in **Table 2-3.**

Table 2: Concentration of heavy metals (mg kg-1 dry weight) in *Piper Betel* Leaf (PBL)

Sampling Station	Mn	Cu	Zn	As	Cr	Cd	Pb
S-01	1.780	2.313	20.405	0.088	0.438	0.091	0.421
S-02	1.842	6.808	19.109	0.082	0.382	0.110	0.318
S-03	0.508	4.024	11.209	0.077	0.330	0.052	0.212
S-04	1.21	4.118	16.236	0.065	0.237	0.065	0.304
S-05	0.505	12.117	8.213	0.110	0.260	0.091	0.568
S-06	1.529	11.825	4.086	0.057	0.615	0.157	0.102
S-07	0.317	1.332	8.605	0.061	0.236	0.079	0.532
S-08	0.709	3.427	10.312	0.092	0.232	0.120	0.413
S-09	1.105	0.241	3.214	0.116	0.308	0.072	0.286
S-10	0.825	0.403	2.625	0.069	0.3415	0.105	0.123
S-11	1.119	2.924	5.189	0.091	0.645	0.039	0.169
S-12	1.124	0.535	4.321	0.108	0.560	0.072	0.703
S-13	0.718	2.015	7.208	0.052	0.418	0.112	0.434
S-14	0.827	2.618	9.005	0.101	0.216	0.079	0.507
S-15	0.356	0.329	6.624	0.062	0.327	0.065	0.291
S-16	1.303	0.810	13.142	0.071	0.210	0.104	0.320
S-17	0.415	0.524	1.419	0.079	0.516	0.052	0.239
S-18	1.015	0.619	0.821	0.056	0.115	0.069	0.345
S-19	0.913	1.518	9.369	0.078	0.529	0.109	0.161
S-20	0.518	2.221	7.218	0.101	0.445	0.049	0.079
S-21	0.227	4.409	8.103	0.109	0.218	0.038	0.287
S-22	0.331	0.752	6.862	0.107	0.214	0.052	0.109
S-23	1.452	7.619	5.575	0.099	0.191	0.105	0.501
S-24	1.509	3.528	5.624	0.082	0.409	0.076	0.209
S-25	1.089	4.718	5.551	0.109	0.334	0.116	0.421
S-26	1.118	3.102	6.576	0.082	0.322	0.103	0.112
S-27	3.627	3.827	11.221	0.095	0.154	0.081	0.208
S-28	2.512	4.418	7.538	0.114	0.210	0.075	0.510
S-29	0.309	1.803	8.132	0.076	0.135	0.070	0.605
S-30	2.518	2.612	5.616	0.079	0.431	0.119	0.201
Mean	**1.110**	**3.250**	**7.971**	**0.086**	**0.333**	**0.084**	**0.323**
Range	**0.227-3.627**	**0.241-12.117**	**0.821-20.450**	**0.052-0.116**	**0.115-0.645**	**0.038-0.157**	**0.079-0.703**

Table 3: Concentration of heavy metals (mg kg-1 dry weight) in Areca Nut (AN)

Sampling Station	Mn	Cu	Zn	As	Cr	Cd	Pb
S-01	2.761	4.513	6.109	0.042	0.342	0.121	0.103
S-02	2.123	4.292	5.125	0.051	0.315	0.105	0.295
S-03	2.541	2.130	5.921	0.037	0.279	0.097	0.265
S-04	1.918	7.659	7.126	0.041	0.413	0.085	0.311
S-05	1.219	5.332	6.093	0.072	0.399	0.125	0.123
S-06	2.505	6.617	2.334	0.096	0.251	0.091	0.198
S-07	3.313	3.561	4.429	0.049	0.360	0.067	0.297
S-08	1.345	4.301	5.281	0.071	0.294	0.082	0.282
S-09	1.612	6.144	6.094	0.09	0.369	0.151	0.351
S-10	3.371	5.301	6.518	0.068	0.522	0.076	0.176
S-11	3.213	5.056	3.429	0.062	0.419	0.112	0.121
S-12	1.830	1.220	3.056	0.057	0.145	0.081	0.293
S-13	1.051	8.312	3.907	0.045	0.505	0.095	0.192
S-14	1.216	3.304	5.256	0.088	0.463	0.089	0.257
S-15	1.371	5.185	1.921	0.032	0.349	0.052	0.284
S-16	1.105	6.122	10.205	0.081	0.629	0.093	0.153
S-17	1.650	4.089	3.372	0.089	0.489	0.127	0.179
S-18	2.183	7.053	5.739	0.056	0.410	0.105	0.312
S-19	1.219	2.304	4.021	0.075	0.389	0.090	0.286
S-20	1.050	2.174	3.720	0.094	0.380	0.0926	0.187
S-21	0.905	8.139	7.292	0.059	0.372	0.067	0.125
S-22	1.328	6.604	6.122	0.087	0.398	0.074	0.207
S-23	2.197	5.222	6.415	0.084	0.723	0.109	0.195

S-24	3.26	3.262	7.110	0.079	0.669	0.058	0.281
S-25	1.620	9.209	1.080	0.062	0.251	0.065	0.253
S-26	1.044	5.305	2.427	0.089	0.450	0.071	0.182
S-27	0.742	5.173	3.114	0.062	0.628	0.084	0.209
S-28	0.495	6.429	9.172	0.059	0.384	0.092	0.188
S-29	1.209	5.605	5.316	0.036	0.562	0.109	0.199
S-30	1.281	4.785	2.952	0.058	0.871	0.142	0.208
Mean	**1.756**	**5.147**	**5.022**	**0.066**	**0.434**	**0.094**	**0.224**
Range	**0.495-3.371**	**1.22-9.209**	**1.08-10.205**	**0.032-0.096**	**0.145-0.871**	**0.052-0.151**	**0.103-0.351**

The average concentration of Mn, Cu, Zn, As, Cr, Cd, and Pb are observed 1.110, 3.250, 7.971, 0.561, 0.333, 0.084 and 0.323 mg kg^{-1} in PBL; 1.756, 5.147, 5.022, 0.081, 0.434, 0.094 and 0.224 mg kg^{-1} in AN. In this study, the decreasing order of heavy metals in PBL and AN are Zn>Cu>Mn>Cr>Pb>As>Cd and Cu>Zn>Mn>Cr>Pb>Cd>As respectively. The mean concentration of Mn, Cu, Cr, and Cd in AN are found comparatively higher than PBL. According to Pearson correlation analysis among the different heavy metals in PBL and AN (**Table 4-5**), a positive inter-element correlation is found between Cu-Cd (at 0.05 significant level) in PBL samples. While, there is no significant positive or negative correlation present among the other heavy metals in both PBL and AN samples. Therefore, the possible sources of contamination are different for individual heavy metals.

Table 4: Pearson correlation among different heavy metals in PBL

Variables	Mn	Cu	Zn	As	Cr	Cd	Pb
Mn	1						
Cu	0.200	1					
Zn	0.236	0.159	1				
As	0.114	0.136	-0.031	1			
Cr	0.000	0.057	-0.144	-0.110	1		
Cd	0.333	0.414*	0.084	-0.278	0.144	1	
Pb	-0.082	0.079	.0120	0.208	-0.319	0.023	1

* *Correlation is significant at the 0.05 level (2-tailed).*

Table 5: Pearson correlation among different heavy metals in AN

Variables	Mn	Cu	Zn	As	Cr	Cd	Pb
Mn	1						
Cu	-0.220	1					
Zn	0.001	0.111	1				
As	-0.123	-0.104	0.006	1			
Cr	-0.080	0.070	0.210	0.129	1		
Cd	-0.047	-0.049	0.092	0.108	0.250	1	
Pb	0.108	-0.191	-0.131	-0.124	-0.246	-0.156	1

The obtained results from this study have been compared with other similar studies (**Table 6**). Hafeman et al., reported that Bangladeshi people consume 5.7-6.3 BQs per day [48]. In another study Ahmed et al., reported the Bangladeshi people, who are living in the UK, usually consume 2-5 BQs per day [49]. In our survey of 300 BQ chewers from Khulna, Bangladesh, we found an average of 8.33 BQs per day with a range of 2–35 per day. Lin et al., and Wu et al., have also reported that people can chew up to 30 BQs per day [50, 51]. Therefore, intake of BQ could be a possible source of heavy metal exposure to the human. At present, only few data are available about the concentration of heavy metals in PBQ and AN. Among them, Al-Rmalli et al., reported that the highest concentrations of As was found in PBL (0.406 mg kg^{-1}) with a mean concentrations of As in BQ of 0.035 mg kg^{-1} whereas the mean concentrations of Cd and Pb were observed 0.028 mg kg^{-1} and 0.423 mg kg^{-1} in BQ [52]. In another study, Rehman et al., reported the mean concentration of Cr, Mn, Cu, Pb, Cd and As was 0.46, 1.14, 2.70, 1.04, 0.058 and 0.021 mg kg^{-1} in PBL; 0.35, 2.98, 9.41, 2.23, 0.128 and 0.030 mg kg^{-1} in AN [53].

Table 6: Comparison of the mean concentration of heavy metals (mg kg^{-1}) in PBL, AN, and BQ with similar studies

Sample Types	Mn	Cu	Zn	As	Cr	Cd	Pb	Reference
Piper Betel Leaf (PBL)	1.110	3.250	7.971	0.086	0.333	0.084	0.323	**Present Study**
Areca nut (AN)	1.756	5.147	5.022	0.066	0.434	0.094	0.224	
Betel Quid (PBL+AN)	**2.866**	**8.397**	**12.993**	**0.152**	**0.767**	**0.178**	**0.547**	
Piper Betel Leaf (PBL)	--	--	--	0.107	--	0.049	0.725	[52]
Areca nut (AN)	--	--	--	0.013	--	0.016	0.102	
Ordinary Betel Quid (OBQ)	**--**	**--**	**--**	**0.035**	**--**	**0.028**	**0.423**	
Piper Betel Leaf (PBL)	1.14	2.70	--	0.021	0.46	0.058	1.04	[53]
Areca nut (AN)	2.98	9.41	--	0.030	0.35	0.128	2.23	

Furthermore, several literatures reported, BQ chewing causes oral cancer [54] and oral sub-mucous fibrosis [55]. Intake of BQs might be responsible for the accretion of heavy metals in the different parts of the human body, which could inhibit the normal physiological functions and lead to kidney, cardiovascular, and bone diseases. Prolonged exposure of Cd can cause pulmonary effects like emphysema, renal effects, bronchiolitis, and alveolitis [56, 57], while Pb toxicity causes joints problems, dysfunction of kidney, reproductive and cardiovascular systems, enduring impairment to the central and peripheral nervous systems, and lessening in hemoglobin formation [58]. 'As' is another toxic metal which generally targets the pulmonary nervous system and responsible for neuropathy, dermatomes, perforation of the nasal septum, skin respiratory cancer. Similarly, Cr also targets the pulmonary systems and causes an ulcer, respiratory cancer, and perforation of nasal septum [59]. Ingestion of excessive copper can cause temporary gastrointestinal distress with symptoms such as nausea, vomiting, and abdominal pain. High levels of exposure to Cu can destroy red blood cells, possibly resulting in anemia [59] while high Zn content may affect growth and reproduction impairment [60].

Table 7: EDI, THQ, HI and TCR of *Piper betel* leaf (PBL), Areca nut (AN) and Betel quid (BQ)

Parameters		Mn	Cu	Zn	As	Cr	Cd	Pb
EDI (mg kg^{-1} BW day^{-1})	PBL	0.00038	0.00111	0.00272	0.00003	0.00011	0.00003	0.00011
	AN	0.00060	0.00175	0.00171	0.00002	0.00015	0.00003	0.00008
	PBL + AN	0.00099	0.00286	0.00441	0.00005	0.00026	0.00006	0.00019
PTDI[a]		**0.14**	**0.5**	**1**	**0.00214**	**0.003**	**0.00066**	**0.00357**
MTDI[b]		**0.14**	**0.1667**	**0.3**	**0.0018**	**0.0028**	**0.0008**	**0.003**
THQ	PBL	0.00270	0.02769	0.00906	0.09770	0.00008	0.00954	0.03145
	AN	0.00427	0.04385	0.00571	0.07498	0.00010	0.01068	0.02181
	PBL + AN	0.00706	0.07155	0.01469	0.17268	0.00017	0.06067	0.05326
HI					0.33964			
TCR	PBL	--	--	--	--	--	4.6E^{-4}	2.7E^{-4}
	AN	--	--	--	--	--	6.0E^{-4}	1.9E^{-4}
	PBL + AN	--	--	--	--	--	1.1E^{-3}	4.5E^{-4}

[a]*Provisional tolerable daily intake values of heavy metals based on the data established by the joint FAO/WHO Expert Committee on Food Additives [61, 62],* [b]*Maximum tolerable daily intake (mg kg^{-1} BW day^{-1}) [61, 63, 64]*

Table 7 summarizes the non-carcinogenic (THQ, HI), and carcinogenic (TCR) health risk of BQ chewers, which were calculated based on equation (2-4). As seen from the data, THQ values of individual heavy metals never exceeded the acceptable guideline value for THQ is ≤ 1.0 [40]. In this regard, Ambedkar and Maniyan concluded, if the value of THQ for individual heavy metal does not exceed its tolerable limit, the food is considered safe for human consumption [65]. Therefore, the THQ analysis suggested that individual heavy metal does not have any possibility of developing non-carcinogenic health risk. Moreover, the combined impact of all metals (HI) was also less than the maximum tolerable limit 1.0, indicating that these concentrations of human exposure to these heavy metal could not pose any adverse effect during BQ chewer's lifetime. The target carcinogenic risks (TCR) of Cd and Pb were calculated which are shown in **Table 7**. In this study, we did not calculate the TCR for As, due to its high carcinogen slope factor value. From a health risk perspective, the environmental quality standards and food safety standards of As may need to be improved [66]. The TCR values of Cd and Pd are 4.6E^{-4} and 2.7E^{-4} for PBL; 6.0E^{-4} and 1.9E^{-4} for AN; 1.1E^{-3} and 4.5E^{-4} for BQ (**Table 7**).

In BQ samples, TCR of Cd is higher than the USEPA unacceptable level of 10^{-4} [39], whereas TCR of Cd and Pd in PBQ and AN samples are found within the acceptable range, indicating no carcinogenic risk from PBQ and AN consumption, but a small concern for Cd-induced carcinogenic risk does exist, particularly from consumption of BQ.

4. Conclusion

In the present study, the concentrations of heavy metals (Cr, Mn, Cu, Pb, Zn, Cd, and As) in PBQ and AN were determined and the probability of developing non-carcinogenic and carcinogenic health risk were assessed. Our finding shows that the EDI values of heavy metals in PBL and AN are lower than the respective PTDI and TDI values. Besides, the non-carcinogenic parameters (THQ and HI) for heavy metals never exceeded the USEPA risk limit (1.0). Similarly, the values of the carcinogenic parameter (TCR) are lower than the USEPA tolerable limit (10^{-4}) suggesting no potential cancer risk from Pb, Cd consumption for PBL and AN, but slight concern for Cd-induced cancer risk through consumption of BQs. Therefore, the present study concludes that the health risks (either carcinogenic or non-carcinogenic) of examined heavy metals in PBL, AN, and BQ are almost safe for BQ chewers but regular monitoring requires to control contamination burden of heavy metals in this sector.

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6. Conflict of Interests

The authors declare no conflict of interest

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