**Trans-Lycopene and β-Cryptoxanthin Intake and Stomach Cancer in Vietnamese Men: A Pilot Case-Control Study**

Khanpaseuth Sengngam¹, Tran Hieu Hoc³, Dao Viet Hang⁴, Le Tran Ngoan¹,⁵*

**Abstract**

**Objective:** To examine the association between dietary intake of Trans-Lycopene and β-Cryptoxanthin and stomach cancer in Vietnamese men. **Methods:** A case-control study including 80 male incident stomach cancer cases and 146 male controls was performed in a general hospital in Viet Nam. A validated semi-quantitative food frequency (SQFFQ) and demographic lifestyle questionnaire were designed, and venous blood samples were collected to determine *H. pylori* status by IgG ELISA. Nutrient intake was converted using the data of SQFFQ and the Nutritive Composition Table of Vietnamese Foods, updated in 2019. The respective associations between Trans-Lycopene and β-Cryptoxanthin intake and stomach cancer were examined using unconditional logistic regression analysis with adjustments for possible cofactors. **Results:** Both Trans-Lycopene and β-Cryptoxanthin intake and stomach cancer showed a significantly inverse association, tertile-3 versus tertile-1, (OR = 0.15, 95%CI: 0.06–0.35, p trend = 0.00) and (OR = 0.34, 95%CI: 0.14–0.79, p trend = 0.02, respectively). For Trans-Lycopene intake stratifying by *H. pylori* status remained the benefit effect against stomach cancer among *H. pylori*-negative participants (OR = 0.15, 95%CI: 0.03–0.69, p trend = 0.02) and *H. pylori*-positive participants (OR = 0.13, 95%CI: 0.04–0.42, p trend = 0.00). **Conclusions:** Both Trans-Lycopene and β-Cryptoxanthin intake showed a strong protective effect against stomach cancer. The findings suggest that these two types of fat-soluble micronutrients would be considered as an anti-cancer therapy for both primary and secondary prevention.

**Keywords:** Stomach cancer- Helicobacter pylori- Trans-Lycopene and β-Cryptoxanthin intake

**Introduction**

Trans-Lycopene and β-Cryptoxanthin are fat-soluble micronutrients and carotenoids and are found in many foods, including some vegetables, fruits, meats, and animal products. At least 700 carotenoids are found in natural foods (Cited by CDC) (CDC, 2008). Carotenoids are believed among the best biological markers for fruits and vegetable intake. The strongest dietary predictors of serum carotenoid concentration are fruits for sources of β-Cryptoxanthin, and tomato products for Trans-Lycopene (CDC, 2008; CDC, 2012). Both vegetables and fruits are protective against many cancer sites(WCRF, 2007). We hypothesized that Trans-Lycopene and β-Cryptoxanthin intake may play an important role to reduce the risk of developing stomach cancer in humans. The present study aimed to examine the association between dietary intake of Trans-Lycopene and β-Cryptoxanthin and stomach cancer in Vietnamese men.

**Materials and Methods**

**Methods**

A case-control study including 80 male incident stomach-cancer cases and 146 male controls was performed in Bach Mai general university hospital located in the city of Hanoi in the North Viet Nam during 2018-2019, Figure 1.

**Cases and hospital controls recruitment**

The incident stomach-cancer cases were first diagnosed as having stomach cancer by histopathological confirmation and underwent surgery to remove the cancer tumor. The male hospital controls were matching to the cases by +/-5-year-old and admitted into the same surgery department and also undergone surgery on to treat for non-cancer tumor. The patients were able to well communicate to answer study questions by face-to-face interviews by the trained data collectors on times before the operating day. Both cases and hospital controls were agreed upon and signed informed consent to participate in the present study.

¹Department of Occupational Health, Hanoi Medical University, Hanoi, Viet Nam. ²National Institute of Public Health, Lao PDR, Viet Nam. ³Department of Surgery, Hanoi Medical University, Hanoi, Viet Nam. ⁴Department of Internal Medicine, Hanoi Medical University, Hanoi, Viet Nam. ⁵Department of Public Health, School of Medicine, International University of Health and Welfare, Japan. *For Correspondence: letngoan@hmu.edu.vn
The control-per-case ratio was preferable to about two. **Exposure assessments**

A validated semi-quantitative food frequency (SQFFQ) and demographic lifestyle questionnaire were designed including 113 questions. Additional 79 questions were added to collect data on cooking methods and pre-clinical examination results. Nutrient intake was converted using the data of SQFFQ and the Nutritive Composition Table of Vietnamese Foods, updated in 2019. The SQFFQ inquired about dietary habits during the previous one year for 85 foods/recipes and frequency in 7 categories: seldom or never intake, 6-11 times/year, 1-3 times/month, 1-2 times/week, 3-4 times/week, 5-6 times/week, and 1-3 times/day. Nutrients intake was estimated following the formulation, based on the Nutritive Composition Table of Vietnamese Foods, updated in 2019, such as:

\[
n = f \times p \times (w/100) \times k / 365
\]

Where “f” was frequency intake during the last year; “p” was the amount of nutrient per 100 gram; “w” was average weight intake of one time or service; “k” was the weighted size of intake amount (small=0.8, medium=1.0, large=1.2). The designed SQFFQ was being validated as having a good characteristic of feasibility, practicality, and reliability in the general populations located in North Viet Nam (Le et al., 2018).

For tobacco smoking, participants were asked about the types of tobacco (cigarette, waterpipe tobacco, others); the average number of tobacco consumed per day during their age of 15-20, 21-15, 26-30, 31-40, 41-50, 51-60, 61-70, and 71+ (if applicable); the duration of smoking (for the current smoker); and the duration of quit smoking (for current and ex-smokers).

To investigate the presence of *H. pylori* infection, 3 ml aliquots of overnight fasting venous blood were collected from both cases and controls. The blood tube was shaken gently several times to mix blood and EDTA-2Na, quickly followed by centrifugation at 3,000 rpm for 20 minutes in a centrifuge with low-temperature control. The anti-*H. pylori* serum IgG titer were tested by enzyme-linked immunosorbent assay (ELISA) based on the sandwich principle using *H. pylori* IgG ELISA kit (RE56381) from IBL International (Hamburg, Germany). The experienced investigators of the Microbiology and Infectious Disease, College of Veterinary Medicine, Vietnam National University of Agriculture performed laboratory work for all plasma samples with the ID of study participants only to avoid information bias and systematic errors in determining the status of *H. pylori* infection.

**Data analysis**

The respective associations between *Trans*-Lycopene and β-Cryptoxanthin intake and stomach cancer were examined using unconditional logistic regression analysis with adjustments for possible cofactors of the age groups (20-49, 50-59, 60-69, ≥70 ages), education levels (primary school, secondary school, high school or higher), BMI (<18.5, 18.5 to <23, 23 to <25, ≥25 kg/m²), tobacco smoking (ever/never smokers), consumption of five common types of vegetables (water spinach, mustard greens, saurupods, Malabar nightshade, cabbage); and *H. pylori* infection status, total calorie intake, and family history of cancer. Sensitive analysis was done for the sub-groups of tobacco smoking (ever/never smokers) and *H. pylori* infection status. All p-values were two-sided, and p ≤ 0.05 (alpha value) was considered to indicate statistical significance.

**Results**

*H. pylori*-positive was 50.00% of cases and 38.36% of controls. Ever tobacco smokers were 88.75% of cases and 73.97% of controls. Normal body-mass-index (18.5 to <23.0) was 52.5% of cases and 50.00% of controls. Less than 6-year education was 18.75% of cases and 12.33% of controls, Table 1.

Both *Trans*-Lycopene and β-Cryptoxanthin intake and stomach cancer showed a significantly inverse association, tertile-3 versus tertile-1, (OR = 0.15, 95%CI: 0.06–0.35, p < 0.001), Table 1.

### Table 1. Characteristics of the Study Population

| Variables          | Stomach cancer | Non-cancer |
|--------------------|----------------|------------|
|                    | n   | %  | n   | %  |
| **Age group (ages)** |     |    |     |    |
| 20-49              | 14  | 17.50 | 47 | 32.19 |
| 50-59              | 24  | 30.00 | 53 | 36.3 |
| 60-69              | 28  | 35.00 | 31 | 21.23 |
| ≥70                | 14  | 17.50 | 15 | 10.27 |
| Total              | 80  | 100.00 | 146 | 100.00 |
| **Education (years)** |     |    |     |    |
| <6                 | 15  | 18.75 | 18 | 12.33 |
| 6-9                | 36  | 45.00 | 64 | 43.84 |
| 10-12              | 15  | 18.75 | 46 | 31.51 |
| >12                | 13  | 16.25 | 17 | 11.64 |
| Unknown            | 1   | 1.25 | 1  | 0.68 |
| Total              | 80  | 100.00 | 146 | 100.00 |
| **BMI (kg/m²)**    |     |    |     |    |
| 18.5 to <23        | 42  | 52.5 | 73 | 50.00 |
| 23 to <25          | 9   | 11.25 | 30 | 20.55 |
| ≥25                | 5   | 6.25 | 18 | 12.33 |
| <18.5              | 24  | 30 | 22 | 15.07 |
| Unknown            | 0   | 0.00 | 3  | 2.05 |
| Total              | 80  | 100.00 | 146 | 100.00 |
| **Lifetime tobacco smoking** |     |    |     |    |
| Never              | 9   | 11.25 | 38 | 26.03 |
| Ever               | 71  | 88.75 | 108 | 73.97 |
| Total              | 80  | 100.00 | 146 | 100.00 |

a, BMI, Body Mass Index (BMI = weight (kg) / height (m²)); b, Classification according to the manufacturer's instructions according to anti-*H. pylori* IgG concentration of the Cut-off Index (COI) quantitative
Table 2. Daily Intake of Trans-Lycopene and Stomach Cancer in Vietnamese Men

|                | Total & | Daily mean intake (µg) | Control | Case | Total | OR (95%CI) | P for trend |
|----------------|---------|------------------------|---------|------|-------|------------|------------|
| Total          |         |                        |         |      |       |            |            |
|                |         |                        | 332.6   | 38   | 38    | 76         | 1          |
|                |         |                        | 752.8   | 47   | 30    | 77         | 0.53 (0.26, 1.08) |
|                |         |                        | 1538.3  | 61   | 12    | 73         | 0.15 (0.06, 0.35) |
|                |         |                        | Total   | 146  | 80    | 226        | 0          |
| Never smoker & |         |                        | 334     | 9    | 5     | 14         | 1          |
|                |         |                        | 793.7   | 10   | 2     | 12         | 0.17 (0.01, 1.97) |
|                |         |                        | 1671.2  | 19   | 2     | 21         | 0.21 (0.02, 2.46) |
|                |         |                        | Total   | 38   | 9     | 47         | 0.18       |
| Ever smoker &  |         |                        | 332.3   | 29   | 33    | 62         | 1          |
|                |         |                        | 745.2   | 37   | 28    | 65         | 0.57 (0.26, 1.23) |
|                |         |                        | 1484.7  | 42   | 10    | 52         | 0.14 (0.05, 0.37) |
|                |         |                        | Total   | 108  | 71    | 179        | 0          |
| H. pylori negative $ |   |                        | 372.5   | 17   | 12    | 29         | 1          |
|                |         |                        | 710.9   | 16   | 8     | 24         | 1.08 (0.28, 4.27) |
|                |         |                        | 1599.5  | 30   | 5     | 35         | 0.15 (0.03, 0.69) |
|                |         |                        | Total   | 63   | 25    | 88         | 0.02       |
| H. pylori positive or equivocal $ |         |                        | 308     | 21   | 26    | 47         | 1          |
|                |         |                        | 771.8   | 31   | 22    | 53         | 0.37 (0.15, 0.91) |
|                |         |                        | 1482    | 31   | 7     | 38         | 0.13 (0.04, 0.42) |
|                |         |                        | Total   | 83   | 55    | 138        | 0          |

OR (95% CI): odds ratio 95% confidence interval. &: Adjusted for age groups (20-49, 50-59, 60-69, ≥70 ages), education levels (primary school, secondary school, high school or higher), BMI (<18.5, 18.5 to <23, 23 to <25, ≥25 kg/m2), tobacco smoking (ever/never smokers), consumption of five common types of vegetables (water spinach, mustard greens, sauropods, Malabar nightshade, cabbage); and H. pylori infection status, total calorie intake, and family history of cancer. &&: Adjusted for age groups (20-49, 50-59, 60-69, ≥70 ages), education levels (primary school, secondary school, high school or higher), BMI (<18.5, 18.5 to <23, 23 to <25, ≥25 kg/m2), tobacco smoking (ever/never smokers), consumption of five common types of vegetables (water spinach, mustard greens, sauropods, Malabar nightshade, cabbage), and H. pylori infection status, total calorie intake, and family history of cancer. $: Adjusted for age groups (20-49, 50-59, 60-69, ≥70 ages), education levels (primary school, secondary school, high school or higher), BMI (<18.5, 18.5 to <23, 23 to <25, ≥25 kg/m2), tobacco smoking (ever/never smokers), consumption of five common types of vegetables (water spinach, mustard greens, sauropods, Malabar nightshade, cabbage), total calorie intake, and family history of cancer;
trend = 0.00) and (OR = 0.34, 95%CI: 0.14–0.79, p trend = 0.02, respectively). For Trans-Lycopene intake stratifying by H. pylori status remained the benefit effect against stomach cancer among H. pylori-negative participants (OR = 0.15, 95%CI: 0.03–0.69, p trend = 0.02) and H. pylori-positive participants (OR = 0.13, 95%CI: 0.04–0.42, p trend = 0.00). By smoking habits, a significant negative association between Trans-Lycopene intake and stomach cancer was observed among ever smokers (tertile-3 versus tertile-1, OR = 0.14, 95%CI: 0.05–0.37, p for trend = 0.00), but no such statistically significant relationship was found among never smokers (OR = 0.21, 95%CI: 0.02–0.46, p for trend = 0.18), Table 2.

For β-Cryptoxanthin intake by H. pylori status, the benefit effect against stomach cancer was not observed among H. pylori-negative participants (OR = 0.51, 95%CI: 0.13–2.02, p trend = 0.35) but that was seen among H. pylori-positive participants (OR = 0.19, 95%CI: 0.06–0.65, p trend = 0.01). By smoking habits, a significant negative association between β-Cryptoxanthin intake and stomach cancer was observed among ever smokers (tertile-3 versus tertile-1, OR = 0.14, 95%CI: 0.04–0.91, p for trend = 0.03), but no such statistically significant relationship was found among never smokers (OR = 0.23, 95%CI: 0.02–2.87, p for trend = 0.26), Table 3.

### Discussion

We observed a strong protective effect against stomach cancer by both two natural micronutrients of Trans-Lycopene and β-Cryptoxanthin intake and this activity strongly remained among H. pylori-positive participants and ever-smoker individuals. The findings suggest that these two types of fat-soluble micronutrients would be considered as an anti-cancer therapy for primary prevention in controlling the occurrence and mortality from stomach cancer.

The present findings are supporting data of Trans-Lycopene and β-Cryptoxanthin regarding their roles of antioxidant activity, anti-inflammatory activity, and
anti-cancer. The carotenoids are fat-soluble micronutrients that are found in many foods of vegetables, fruits, meats, and animal products. Six major carotenoids are found in human blood including alpha-carotene, beta-carotene, Beta-cryptoxanthin, lutein, Trans-lycopenes, and zeaxanthin (Li et al., 2021). Carotenoids are believed among the best biological markers for fruit and vegetable intake. Trans-Lycopene, molecular formula C40H56, is a natural strong antioxidant to protect against cancer. Major Trans-Lycopene is obtained primarily from tomato and other pink orange or red fruits and vegetables mainly including guava, papaya, peaches, pink grapefruit, red carrots, gac melons, watermelons, apricot, cranberry. Academic reports have found that lycopene has antioxidant, anti-inflammatory, anti-carcinogenic (Li et al., 2021). However, the association between dietary lycopene intake and stomach cancer is inconclusive (Yang et al., 2013), (Ito et al., 2005). Beta-cryptoxanthin as provitamin A is an essential nutrient supporting the growth development and maintenance of the immune system. Beta-cryptoxanthin can do antioxidant activity, anti-inflammatory activity, and anti-cancer (Burri et al., 2016). Stomach cancer (or gastric cancer) is the fifth most frequent cancer incidence and the third leading cancer death globally (GLOBOCAN 2018) (Bray et al., 2018). In Asia, stomach cancer is the third most common cancer and the second most common cause of cancer death with the highest incidence rates were observed in the Eastern and Southeastern regions (Colquhoun et al., 2012). The present findings add data to promote cancer prevention based on a healthy diet and intake of natural food rich in Trans-Lycopene and β-Cryptoxanthin. The present study has certain limitations of the absent data in women and limited sample size as a pilot case-control study in the low- and middle economy country of Viet Nam. The other possible limitation is that there were available hospital controls only and a recall of dietary habits might be a low accuracy and completeness of food frequency intake. Despite these limitations, the results support the hypothesis of Trans-Lycopene and β-Cryptoxanthin intake may play an important role to reduce the risk of developing stomach cancer in humans.

**Author Contribution Statement**

Conceptualization: KS, THH, DVH, LTN. Data curation: NTL. Formal analysis: NTL. Funding acquisition: NTL. Methodology: NTL, KS. Project administration: DVH, NTL. Visualization: THH, NTL. Writing - original draft: NTL. Writing - review & editing: DVH, NTL.

Acknowledgments

We used a research grant No.: 18/FIRST/1a/HMU, under the Project: “Fostering Innovation through Research, Science and Technology”. It is a part of a Ph.D. student research thesis that was approved by the Hanoi Medical University academic committee dated 13 July 2018 and by the Ethical Committee No. of approval 3918/ HMUIRB dated 25 December 2018. We thank the study participants and their family members for their fruitful time and support in providing data of smoking, diet, health history, blood sample, and other information of demographic lifestyles for the present study. We have appreciated all staff of the health facilities involved in the present research and management.

**Available data**

The original database will be available upon appropriate request.

**Conflict of interest**

There are no conflicts to disclose

**References**

Bray F, Ferlay J, Soerjomataram I, et al (2018). Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin, 68, 394-424.

Burri BJ, La Frano MR, Zhu C (2016). Absorption, metabolism, and functions of beta-cryptoxanthin. Nutr Rev, 74, 69-82.

CDC (2008). National Report on Biochemical Indicators of Diet and Nutrition in the U.S. Population 1999–2002. In Fat-Soluble Vitamins & Micronutrients: Vitamins A and E and Carotenoids; Available at https://www.cdc.gov/nutritionreport/pdf/nutrition_report.pdf, CDC (ed). Center for Diseases Control, pp 35-59.

CDC (2012). Second National Report on Biochemical Indicators of Diet and Nutrition in the U.S. Population 2012. In Fat-Soluble Vitamins & Micronutrients: Vitamins A and E and Carotenoids; Available at https://www.cdc.gov/nutritionreport/pdf/nutrition_book_complete508_final.pdf, CDC (ed). Center for Diseases Control, pp 35-59.

Colquhoun A, Arnold M, Ferlay J, et al (2012). Global patterns of the cardia and non-cardia gastric cancer incidence in 2012. Gut, 64, 1881-8.

Ito Y, Kurata M, Hioki R, et al (2005). Cancer mortality and serum levels of carotenoids, retinol, and tocopherol: a population-based follow-up study of inhabitants of a rural area of Japan. Asian Pac J Cancer Prev, 6, 10-5.

Le TN, Le XH, Pham VP, et al (2018). Reproducibility of a designed semi-quantitative food frequency questionnaire in general populations in North Viet Nam. Southeast-Asian J Sci, 6, 188-97.

Li N, Wu X, Zhuang W, et al (2021). Tomato and lycopene and multiple health outcomes: an umbrella review. Food Chem, 343, 128396.

WCRF (2007). Food, Nutrition, Physical Activity, and the Prevention of Cancer: a Global Perspective. World Cancer Research Fund / American Institute for Cancer Research: Washington DC AICR.

Yang T, Yang X, Wang X, Wang Y, Song Z (2013). The role of tomato products and lycopene in the prevention of gastric cancer: a meta-analysis of epidemiologic studies. Med Hypotheses, 80, 383-8.