RISK FACTORS AND GENETIC POLYMORPHISMS OF GSTM1, GSTT1 AND CYP1A1 IN DEVELOPMENT OF ORAL CANCER IN AN INDONESIAN POPULATION

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ABSTRACT

Introduction

Cancer is one of the foremost causes of death after infections and heart diseases in all societies and is the fatal leading disease in the world. Oral cancer is the sixth most common cancer worldwide and the most common in South East Asia. The geographic pattern and trends in incidence of oral cancer vary widely between countries and geographical areas of the world. This difference in distribution of oral cancer worldwide is influenced by their risk factors. The major risk factors of oral cancer which have already been established are tobacco use, alcohol drinking and betel quid chewing. Furthermore, epidemiology studies have also implicated other factors such as genetic susceptibility and the role of diet. Jakarta, the capital city of Indonesia is the most populous place in Indonesia. It is inhabited with more than 50% of Indonesia's population. Jakarta city comprises of inhabitants of various ethnicity and practicing different habits. To date no studies have been undertaken to determine the risk factors including genetic polymorphism of metabolizing gene (GSTM1, GSTT1 and CYP1A1) in the development of oral cancer in the Jakarta population.

Objective/Aim

This study aims to identify and determine the risk factors including genetic polymorphisms in GSTM1, GSTT1 and CYP1A1 in the development of oral cancer in a Jakarta population, Indonesia.

Material and Method

This is a cross sectional study with case-control design involving hospital-based subjects matched for age and sex. Two hundred forty three subjects participated in this study from 5 selected hospitals which represent each district of Jakarta, with a 1:2 ratio of cases and

controls. This study consists of two parts. The first part consists of data collection on the subjects' socio demographic characteristics, risk habits (smoking, alcohol consumption and betel quid chewing) and dietary pattern using a structured questionnaire. The second part consists of laboratory work on blood samples of cases and controls to assess the polymorphism of GSTM1, GSTT1 and CYP1A1 by means of PCR and Restriction Fragment Length Polymorphism (RFLP). The data was analysed using conditional logistic regression (STATA 8) and factor analysis (SPSS12) for dietary pattern.

Results

In terms of socio demographic characteristic, 42.3% of oral cancer cases were more than 49 years of age (mean 47.4 years ± 12.4), males being more frequently affected than females with a ratio of 6:4 and the number increases with increasing age-group. Ninety percent of cases were married, and the most affected ethnic group was Deutro Melayu (87.7 %).

The most common habit practiced among the cases (54.2 %) and controls (49.3%) was smoking, either as a single habit or in combination with alcohol and betel quid chewing. Smokers accounted for 55.6% of the cancer cases whilst there was an almost similar proportion of smoker and non smoker among the controls (48.8% cases vs 51.2% controls). Among cases and controls, the majority had smoked for more than 10 years. *Kretek* was the most preferred type of cigarettes for both cases and controls. Almost all smoking factors (number of sticks per day, duration of smoking, and type of tobacco and pack-years of exposure of tobacco) displayed higher risks of oral cancer by two to three times after allowing for confounding factors.

Alcohol drinking habit was practiced by less than 10% of case and control groups (8.6% and 4.3%, respectively). The crude OR showed that alcohol drinking habit did not

contribute to the risk of oral cancer ($p>0.250$). The exception was that those who consumed wine had 11 times higher risk of oral cancer.

Similarly, betel quid chewing habit was also practiced by a very small number of case (7.4%) and control subjects (1.8%). The findings showed that duration of chewing and number of quid per day did not significantly contribute to the risk of oral cancer. However, current betel quid chewer and quid combination of betel leaf, tobacco, areca nut, and lime were significantly associated ($p<0.05$) with increasing risk of oral cancer (OR 5.32, 95% CI 1.03-27.52; OR 4.19, 95% CI 1.05-16.82 respectively).

The genetic polymorphism of GSTM1, GSTT1 and CYP1A1 assay showed that there was no statistically significant difference observed between cases and controls. The polymorphism of GSTM1, GSTT1 and CYP1A1 either singly or in combination did not have association with risk of oral cancer even in the smoker, non smoker as well as among betel quid chewer and non chewer in this study.

Analysis of dietary pattern found four factors retained from factor analysis, referred to as preferred food, combination, chemical related and traditional that were associated with oral cancer after adjusting for smoking, alcohol and betel quid chewing. The preferred food consisted of fast food, fermented food, canned food, snacks high in fat and sugar, cooked and raw vegetables, and seafood showed increased risk of oral cancer by two times (OR 2.17, 95% CI 1.05-4.50, $\chi 2$ trend 5.446, $p<0.05$). The "chemical related" pattern (which is consisted of MSG and processed food) also showed an increase risk (OR 2.85, 95% CI 1.34-6.05). The "combination" pattern (consisted of meat, dairy product and fruit) displayed protective effects in relation to oral cancer (adjusted OR 0.46, 95% CI 0.23-0.91, $\chi 2$ trend 7.335, $p<0.01$). Finally, "traditional" pattern (consist of drinks and carbohydrate)

showed an increased of risk by two-fold (OR 2.04, 95%CI 1.01-4.41, $\chi 2$ trend 5.649, $p<0.05$). The fast food was the most common food type consumed in this study (Communality : 0.818)

Conclusion

Smoking and betel quid chewing were found to be risk factors for oral cancer in the Jakarta population. In contrast genetic polymorphism of GSTM1, GSTT1 and CYP1A1 did not contribute to the development of oral cancer in this study. The dietary pattern referred to as "preferred food", "chemical related" and "traditional" were found to be risk factors of oral cancer whereas the "combination" pattern had protective effect of oral cancer in this study.

Recommendations

Future research is recommended to include a bigger sample size and greater cross section that constitute the Indonesian population and using community controls. Investigation involving nutritional content of food consumed is also advocated.

LIST OF CONTENT

		PAGE
COVER PAGE		i
ABSTRACT		ii
ACKNOWLEDGEMENTS		vi
LIST OF CONTENTS		ix
LIST OF APPENDICES		xv
LIST OF FIGURES		xvi
LIST OF TABLES		xvii
ABBREVIATIONS		xvii
CHAPTER 1	INTRODUCTION	1
CHAPTER 2	PURPOSE OF STUDY	7
CHAPTER 3	LITERATURE REVIEW	9
3.1	Terminology and classification of oral cancer	9
3.2	Epidemiology of oral cancer	13
3.2.1.	Global epidemiology of cancer	13
3.2.2.	Incidence of oral cancer in the world	17
3.2.3.	Incidence of oral cancer in developed countries	18
3.2.4.	Incidence of oral cancer in developing countries	19
3.2.5.	Oral cancer mortality	21
3.2.6.	Gender distribution of oral cancer	22
3.2.7.	Age distribution of oral cancer	23
3.2.8.	Ethnic distribution of oral cancer	24

3.3 Clinical and Pathologic Characteristic of Oral Cancer and Precancer 25

3.3.1 Clinical presentation of oral cacner 26

3.3.1.1 White lesion 26

3.3.1.2 Red lesion 26

3.3.1.3 Ulcerated lesion 26

3.3.1.4 Exophytic lesion 27

3.3.2 Clinical and Pathologic Characteristic of Precancer (Potentially Malignant Lesions/Conditions) 27

3.3.2.1 Leukoplakia 29

3.3.2.2 Erythroplakia 30

3.3.2.3 Malignant potential of precancer and other lesions/conditions 30

3.3.3 Histopathology of oral cancer 32

3.3.4 Sites of cancer 33

3.3.4.1 Tongue cancer 33

3.3.4.2 Floor of the mouth 35

3.3.4.3 Buccal mucosa and lip 36

3.3.4.4 Gum and palate 37

3.4 Carcinogenesis and Risk Factors 38

3.4.1 Carcinogenesis 38

3.4.2 Risk Factors 40

3.4.2.1 Tobacco use as a risk factor

3.4.2.1.1 Carcinogenicity of tobacco 45

3.4.2.1.2 to oral tissue carcinogenesis 47

3.4.2.1.3 Tobacco products 49
3.4.2.2 Alcohol drinking as a risk factor 50
3.4.2.2.1 Variation of content of alcohol beverages/ drinking frequency and risk of oral cancer 51
3.4.2.2.2 Carcinogenicity of alcohol beverages and possible mechanism of oral cancer 54
3.4.2.3 Quid chewing as a risk factor 58
3.4.2.3.1 Tobacco quid/smokeless tobacco 59
3.4.2.3.2. Geographic distribution and variation 62
3.4.2.3.3 Betel quid constituents and effect of areca nut of quid chewing 64
3.4.2.4 Genetic susceptibility 66
3.4.2.5 Diet and nutrition as risk/protective factors 72
3.4.2.6 Viral, Candida infection and other factors 78
3.4.1.6.1 Virus 78
a. Human Papilloma Virus 78
b. Ebstein Barr-Virus 81
3.4.1.6.2 Candida albicans 82
3.4.1.6.3 Other risk factors 82
3.5 The population of Indonesia 84
3.4.2 Population characteristic 84
3.4.3 Incidence/prevalence of oral cancer 86
3.4.4 Risk habits 86

3.5.3.1 Tobacco smoking 86

3.5.3.2 Betel quid chewing 90

3.5.3.3 Alcohol drinking 91

CHAPTER 4 MATERIALS AND METHODS 92

4.1 Study design 92

4.2 Conduct of study 92

4.3 Recruitment of subjects 92

4.3.1. Cases 92

4.3.2. Controls 93

4.3.3. Number of subjects in study population 94

4.4 Questionnaire survey 95

4.4.1. Components in questionnaire survey 95

4.4.1.1. Risk factor 95

4.4.1.1.1. Smoking 95

4.4.1.1.2. Alcohol consumption 96

4.4.1.1.3. Quid chewing 96

4.4.1.1.4. Dietary habit 97

a. Daily food pattern 97

b. Food preparation method 97

c. Food frequency 98

4.4.2. Validation of questionnaire 99

4.4.3. Pre-test of questionnaire 100

4.4.4. Conduct of questionnaire 100

4.5. Laboratory procedure for genetic polymorphism 101

4.5.1. Tissue collection and storage 101

4.5.2. Blood collection and DNA isolation 102
4.5.2.1. Genotyping assay of GSTM1 and GSTT1 102
4.5.2.2. Genotyping assay of CYP1A1 106
4.6 Research ethics 109
4.7 Data analysis 109

CHAPTER 5 RESULTS 113
5.1 Subjects recruited for study 113
5.2 Distribution of cases and controls by centers 114
5.3 Distribution of cases by anatomic sites 114
5.4 Characteristic of study population 115
5.5 Test reliability test of scales 116
5.6 Distribution of cases and controls according to risk habit 117
5.6.1 Smoking habit and risk of oral cancer 119
5.6.2 Alcohol habits and risk of oral cancer 122
5.6.3 Betel quid chewing habit and risk of oral cancer 123
5.6.4 Genetic polymorphism and risk of oral cancer 125
5.6.5 Dietary habits and risk of oral cancer 133
5.6.5.1 Factor analysis of dietary pattern 134

CHAPTER 6 DISCUSSION 139
6.1 Limitation of study 139
6.2 Hypothesis 142
6.3 Socio demographic characteristic of Jakarta population 143
6.4 Habits in Jakarta population and risk of oral cancer 145
6.4.1 Smoking habit and risk of oral cancer 145

6.4.2 Alcohol drinking habit and risk of oral cancer 148

6.4.3 Betel quid chewing habit and risk of oral cancer 150

6.4.4 Genetic polymorphism and risk of oral cancer 153

6.4.5 Dietary habits and risk of oral cancer 158

CHAPTER 7 CONCLUSION AND RECOMMENDATIONS 166

7.1 Conclusion 166

7.1.1 Tobacco smoking 167

7.1.2 Alcohol drinking 167

7.1.3 Betel quid chewing 168

7.1.4 Genetic polymorphism of GSTM1, GSTT1 and CYP1A1 168

7.1.5 Dietary pattern 168

7.2 Recommendation 169

BIBLIOGRAPHY 171

APPENDICES 196

LIST OF FIGURES

		PAGE
1. Figure 3.1 :	Estimated Numbers of New Cancer Cases (Incidence) and Deaths (Mortality) in 2002.Data shown in thousands for developing and developed countries by cancer site and sex	16
2. Figure 3.2 :	Multistep carcinogenesis: initiation, promotion and progression	39
3. Figure 3.3 :	Indonesia Island	85
3. Figure 4.1 :	Flowchart of data collection	94
4. Figure 5.1 :	Sites of oral cancer	115
6. Figure 5.2 :	Distribution of cases and control with and without habit	118
7. Figure 5.3 :	Distribution of cases and controls according to smoking status and duration of smoking	119
8. Figure 5.4 :	Distribution of cases and controls according to number of stick smoked per day and types of tobacco used	120
9. Figure 5.5 :	A Multiplex PCR reaction of GTSM1 and GSTT11 polymorphism	126
10. Figure 5.6 :	CYP1A1 products (370 bp).	126
11. Figure 5.7 :	Restriction Fragment Length Polymorphism (RFLP) for CYP1A1 using Nco1enzyme.	126
12. Figure 5.8 :	Scree plot showing *eigenvalues* for 15 components, in the factor extraction of data obtained from food frequency questionnaire	136

LIST OF TABLES

		PAGE
1. Table 3.1 :	Incidence and Mortality by Sex and Cancer Site Worldwide in year 2002	15
2. Table 3.2 :	Some common forms of oral smokeless tobacco	60
3. Table 5.1 :	Distribution of cases and controls by hospital location	114
4. Table 5.2 :	Distribution of cases and controls by socio demographic variables	116
5. Table 5.3 :	Normality and reliability test of scales	117
6. Table 5.4 :	Risk habits in study population	118
7. Table 5.5 :	Smoking characteristics and risk of oral cancer	121
8. Table 5.6 :	Adjusted OR for smoking	122
9. Table 5.7 :	Alcohol consumption status and risk of oral cancer	123
10. Table 5.8 :	Betel quid chewing status and risk of oral cancer	124
11. Table 5.9 :	Adjusted OR for betel quid chewing status and risk of oral cancer	125
12. Table 5.10 :	Genetic polymorphism and risk of oral cancer	128
13. Table 5.11 :	Distribution and risk of genetic polymorphism among smoker	129
14. Table 5.12 :	Distribution and risk of genetic polymorphism among Non-smoker	130
15. Table 5.13 :	Distribution of genetic polymorphism among quid chewer	131
16. Table 5.14 :	Distribution of genetic polymorphism among non quid chewer	132
17. Table 5.15 :	Frequency distribution of dietary habit in the study population	134
18. Table 5.16 :	Method of food preparation and risk of oral cancer in the study population	135
19. Table 5.17 :	Food group factor loadings for the four dietary patterns identified in the study population	137
20. Table 5.18 :	Odds ratio and 95% CI for oral cancer, in approximate tertile for food groups defined by factor analysis	138

ABBREVIATION

bp	: base pair
BMDP	: BioMeDical Package (statistic software)
CYP	: Cythocrome P450
DNA	: Deoxyribonucleic acid
dNTPs	: deoxyribonucleotide triphosphate
EDTA	: Ethylene Diamine Tetraacetic Acid
FFQ	: Food Frequency Questionaire
GSTM	: Gluthathione S transferase mu
GSTT	: Glutathione S transeferase tetha
KMO	: Kaiser-Meyer-Olkin (Measure of Sampling Adequacy)
MgCL	: Magnesium Chloride
mL	: milliliter
mM	: milliMolar
OR	: Odds Ratio
PCR	: Polymerase Chain Reaction
RFLP	: Restriction Fragment Lenght Polymoprhism
SCC	: Squamous Cell Carcinoma
95% CI	: 95 % Confidence Interval