

Assessment of Omega-3 Fatty Acid Food Intakes in Online Motorcycle Taxi Drivers

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Keywords:

Active smokers, food Frequency Questionnaire, omega-3, online motorcycle taxi drivers, 24- hour recall.

ABSTRACT

Online motorcycle taxi drivers have risk of being exposed to vehicle air pollution, especially with smoking habits. Cigarette smoke can increase lipid peroxidation from PUFA (Polyunsaturated Fatty Acid) by triggering oxidative stress. Low concentrations of omega-3 fatty acids cause dysfunction in dopaminergic system associated with smoking dependence and craving. The purpose was to assess omega-3 fatty acid food intakes in online motorcycle taxi drivers. The research design was case-control method. Measurement of omega-3 intake was using FFQ and 24-hour recall. This research was conducted in Kali Rungkut, Surabaya City, in May-August 2020. The data analysis method used in this research was descriptive analysis. This research involved 49 respondents in the Rungkut area of Surabaya City, it can be concluded that they were not consuming enough food that contains omega-3. The results of omega-3 intake showed that all respondents were in the category of less intake of foods containing omega-3 (<1,600 mg) and average total intake of foods containing omega-3 was 226.47 mg. The most consumed omega-3s by respondents were eggs (an average of 19.46 mg/day) and chicken (an average of 10.74 mg/day), both of which contain low omega-3. Meanwhile, based on data collection per day with a 24-hour dietary recall, it was known that the average daily consumption of respondents is only 249.05 mg. Therefore, it was necessary to further analyze the processing pattern of the food consumed and the factors that influence their diet.



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1. INTRODUCTION

The main sources of air pollution come from various human activities such as industry, transportation, offices and housing. These various kinds of activities are the biggest contribution from air pollutants that are discharged into the free air. The impact of air pollution can cause a decrease in air quality which will have a negative impact on human health [1]. In urban areas, especially in Surabaya, which is the largest metropolitan city in the province of East Java, the activities of its citizens really need fast access, especially

transportation. In today's modern era, people tend to choose fast and practical transportation, one of which is an online motorcycle taxi driver. This can have a high risk of decreased lung function, due to breathing air which is of bad quality for health from exhaust emissions from various motorized vehicles on the road [2], [3]. Decreased lung function can also be affected by smoking. Ninety percent of online motorcycle taxi drivers take advantage of their spare time while waiting for passengers by smoking cigarettes [4], [5]. Cigarettes consist of aerosols and gases, have thousands of chemical components and contain many toxins and are carcinogens. Some of the chemicals contained in cigarettes include nicotine, tar, carbon monoxide and various heavy metals [6], [7]. Nicotine is a drug in the form of tobacco that causes addiction, and nicotine dependence was an indirect root cause for decreased lung function of smokers and showed a gradual decrease in FEV1 with an increase in nicotine dependence [8], [9]. Cigarette smoke is a free radical obtained from exogenous sources. Cigarette smoke is also the main factor affecting the increase in free radicals in the body. An increase in the number of free radicals in the body will trigger oxidative stress and stimulate peroxidation in cells, which can cause damage and death to body cells [10], [11]. Cigarette smoke as free radicals increases Reactive Oxygen Species (ROS) which will result in oxidative stress in the lungs. An imbalance of oxidants and antioxidants in the body causes oxidative stress and triggers an inflammatory reaction in the alveolar tissue. The inflammatory process results in active lung macrophages and neutrophil infiltration which then causes inactivation of the α -AT1 antiprotease, as a pulmonary protease inhibitor in producing pulmonary elastase [12- 14]. This can damage the protein structure of the lung, through the destruction of the alveolar septum so that the elasticity of the lung parenchyma tissue is disrupted and causes changes in the structure and function of the lung tissue [14], [15].

Cigarette smoke can increase lipid peroxidation from PUFA (Polyunsaturated Fatty Acid) by triggering oxidative stress. Low concentrations of PUFA will affect neurotransmission in the central nervous system, namely dopaminergics, which will increase nicotine addiction, thereby inhibiting smoking cessation efforts [16], [17]. Previous research conducted by [18] stated that smokers consume less fish rich in omega-3 fatty acids compared to nonsmokers according to lower levels of DHA (docosahexaenoic acid) and EPA (eicosapentaenoic Acid). The importance of omega-3 intake in smoking, because it can normalize the dopaminergic system so as to reduce addiction. Omega-3 fatty acids were effective in significantly reducing smoking cravings. Cigarettes have a high concentration of free radicals and oxidants that induce systemic oxidative stress and lipid peroxidation, thereby affecting levels of omega-3 PUFAs, particularly EPA and DHA. Smoking increases the bioavailability of long-chain fatty acids from plasma and results in decreased levels of serum omega-3 fatty acids. The brain is susceptible to oxidative stress due to high metabolic activity and susceptibility of PUFAs to free radical attack, so smoking can also reduce levels of omega-3 PUFAs in brain tissue. The long-chain omega-3 PUFAs (EPA and DHA) are important components of cell membranes and as a major part of brain tissue that play a key role in brain function and nerve transmission. Deficiency of omega-3 PUFAs is associated with mesocorticolimbic dysfunction in the dopamine pathway which triggers the release of dopamine, leading to nicotine addiction [16- 19].

Omega-3s have beneficial effects on chronic inflammatory diseases including chronic obstructive pulmonary disease (COPD), asthma, rheumatoid arthritis, and inflammatory bowel disease [20]. Higher omega-3 intake and fish consumption also predict better lung function in smokers and ex-smokers with EPA and DHA acting as antioxidants and reducing oxidative stress [18], [21]. Epidemiological studies show EPA and DHA are the main omega-3 PUFAs found in fish, fish oil and fish oil supplements [22- 24]. In addition, omega-3s can be obtained easily from foods such as salmon, sardines, seafood, and others [25]. According to Recommended Nutritional Adequacy Rate for Indonesian People, concerning the recommended nutritional adequacy rate, for omega-3s of 1.6 g/day in men ≥ 18 years [26]. The pattern of omega-3 intake can be determined by using the FFQ (Food Frequency Questionnaire). The FFQ is a method

of assessing food consumption. The FFQ method is a semi-qualitative method, in which information about the food ingredients consumed is only in the form of names, while the amounts are not clearly differentiated. Every subject who states that they often consume certain foods and drinks, does not always have to be further broken down into the size and portion consumed. The FFQ method only requires data that certain types of food are often or not frequently consumed and their frequency [27- 31]. Previous research conducted by [31] showed that the FFQ can be used to categorize omega-3 fatty acid status. The data collection omega-3 intake in a day is reinforced with a 24-hour recall method, a structured interview intended to capture detailed information about all foods and beverages containing omega-3 [28], [29]. The type of food consumed by a person is strongly influenced by a person's socioeconomic status, cause lower food expenditure is likely to be a key contributor to less-healthy food choices [31]. The purpose of this study was to assess omega-3 fatty acid food Intakes in online motorcycle taxi drivers.

2. METHODS

2.1 Design

The research design used in this study was an observational study, with a case-control method. Measurement of omega-3 intake was carried out using the FFQ and 24-hour recall. This research will be conducted in May- August 2020. This research was conducted in Kali Rungkut, Surabaya City. This research had received ethical permission from the University of Surabaya Ethics Committee Number 016-OL/KE.VII/2020.

2.2 Research Variabel

The variable in this study was the intake of foods containing omega-3. Online motorcycle taxi drivers were public transportation drivers in Indonesia in the form of motorbikes rented by passengers. Orders and payments can be made via the application. An active smoker was someone who has smoked more than 100 cigarettes in their lifetime, and was still consuming cigarettes in the past 28 days. A non-smoker was someone who has never smoked or smoked less than 100 cigarettes in his lifetime [32].

Omega-3 intake was the intake consumed by respondents daily using the FFQ. Researchers will interview respondents with the FFQ which contains the frequency of meals, the number of times the food has been consumed per day, week, month or never. Regular consumption of fish will protect the lungs of smokers by targeting a daily intake of about 1 gram per day of fish or supplements [33]. Many sources of omega-3 were found in salmon, seafood, tuna, fish oil supplements, eggs, soybean oil, canola oil, and others [34], [35]. The way to calculate the omega-3 content was to convert the portion size then look at the list of foods that contain omega-3, then calculate the omega-3 content in food multiplied by the frequency of food from the respondent. The calculation result of the omega-3 content of all foods containing omega-3 in units of milligrams. After that it was compared with the nutritional adequacy rate (RDA) of omega-3 in Indonesia, where if 1600 mg is categorized as sufficient, if <1600 mg is categorized as insufficient [27].

2.3 Population and Sample Research

The population in this study were online motorcycle taxi drivers operating in the Kali Rungkut area of Surabaya City. The sample in this study were online motorcycle taxi drivers operating in the Kali Rungkut area of Surabaya City who met the following criteria: (1) Male; (2) ≥ 18 years old; (3) Active smokers (smoke ≥ 1 cigarette/day); (5) Working 6-8 hours/day; (6) Didn't not have chronic lung function disorders such as asthma, COPD (chronic obstructive pulmonary disease), and respiratory infections; and (7) Not eating vegetarian or being on a certain diet. The sampling technique used purposive sampling and snowball sampling.

2.4 Method of collecting data in Statistics & Data analysis method

The instrument used in this study used a FFQ consisting of 2 parts, namely: demographic data and consumption patterns of food intake containing omega-3 (shrimp, fish sardines, anchovies, catfish, fish salmon, tuna, lobster, crab, shells, eggs, meat chicken, beef, lamb) (Table 1). Preparation for making a questionnaire with several steps as follows: (1) Find list of foods containing omega-3 based on foods that were often consumed by Indonesians [28], [36], [37].

Table 1: List of Composition of Food Intake Containing Omega-3 [28], [36], [37]

The type of food consumed	gram	Omega-3 content (mg/serving)
Eggs	63	22
Chicken meat	100	19
Catfish	85	151
Shrimp	85	267
Beef	100	22
Sardines	57	556
Lamb	100	18
Anchovies	57	1,165
Shellfish	85	241
Crab	85	351
Lobster	100	84
Salmon	85	1,825
Tuna	85	228

The frequency of eating can be calculated in terms of the number of times the food has been consumed each day, week, month or never. This was the first time conducting a preliminary study on online motorcycle taxi drivers who are not research respondents to find out the food on the list that has been made available or often consumed in the area around the research location. This study was conducted by interviewing 20 respondents. Foodstuffs that were consumed by less than 10% of the respondents because they have never been consumed or are not usually consumed are excluded from the food list. Then carry out the final stages or finalization for a list of foods that will be used in the FFQ [29], [30], [38]. After carrying out the final stages or finalizing the FFQ, data collection can be carried out on respondents who had met the criteria. From the interview data, it was processed by estimating the portion size of the food consumed by the respondent into weight (grams) to be able to get the weight of the food consumed in grams/day. After counting all the food items consumed, the known weight consumed in a day (grams/day) was recalculated by looking at the list of foods containing omega-3 and then multiplying by the frequency of meals of the respondent. The data analysis method used in this research was descriptive analysis. Descriptive analysis was method used to describe or describe the collected data without intending to make general conclusions or generalizations.

3. RESULTS

The research design used in this study was an observational study, with case-control method. Measurement of omega-3 intake was carried out using FFQ and 24-hour dietary recall. At the location there were 60 respondents of online motorcycle taxi drivers who met the criteria and only 49 respondents were willing to take part in this study. The rest, as many as 11 respondents, online motorcycle taxi drivers had several reasons so that they were not willing to take part in this research.

The average age being 35.49 years. The lowest age was 21 years, while the highest age was 56 years. Of the 49 respondents, 14 of 49 consumed 1-10 cigarettes per day, 32 of 49 consumed 11-20 cigarettes per day, and 3 of 49 consumed >20 cigarettes per day. Most of the respondents included based on the Brinkman

index were included in mild smokers level (32 of 49). In addition, education level most of the respondents are senior high school (36 of 49) (Table 2).

Table 2: Frequency Distribution of Respondents

Characteristics		Frequency (n=49)	Percentage (%)
Age (years)	18-25	9	18.40
	26-35	17	34.70
	36-45	17	34.70
	46-55	4	8.20
	56-65	2	4.10
Number of Cigarette per day [39]	1-10	14	28.57
	11-20	32	65.31
	>20	3	6.12
Duration of smoking [39]	<10	17	34.69
	10-20	23	46.94
	<20	9	18.37
Brinkman Index [40]	Mild (0-199)	32	65.3
	Moderate (200-600)	15	32.65
	Severe (>600)	2	4.08
Level of education	Elementary school	3	6.10
	Junior high school	2	4.10
	Senior high school	36	73.50
	Bachelor degree	8	16.30

The results of omega-3 intake showed that all respondents were in the category of less intake of foods containing omega-3 (<1,600 mg). The average total intake of foods containing omega-3 was 226.47 mg, with minimum value was 9.43 mg and maximum value was 1,116.34 mg. The type of food containing omega-3 that was consumed the most was chicken eggs by all respondents (average: 19.46 mg/day), although when calculated from the omega-3 content in eggs only 22 mg/serving of 63 g eggs. The least type of food was crab in 5 of the 49 respondents (average: 16.38 mg/day) (Table 3).

Table 3: Profile Distribution of Foods Containing Omega-3 by FFQ

The type of food consumed	Frequency (n=49)	Mean (mg)	SD	Minimum (mg)	Maximum (mg)
Chicken eggs	49	19.46	15.41	0.73	66.00
Chicken meat	47	10.74	4.76	0.63	30.40
Catfish	33	16.36	16.31	2.37	71.05
Shrimp	30	107.04	142.19	8.90	801.00
Beef	26	3.53	4.32	0.73	12.57
Sardines	17	192.19	224.64	18.53	556.00
Lamb	14	0.60	1.20	0.60	0.60
Anchovies	12	156.51	221.92	9.43	613.16
Shellfish	10	55.66	50.21	8.03	103.29
Crab	5	16.38	10.46	11.7	35.1

Meanwhile, based on data collection per day with a 24-hour dietary recall, it was known that the average daily consumption of respondents is only 249.05 mg, with the minimum value being 0 mg, the maximum value was 1069.43 mg. The profile of food consumed per day containing omega-3 can be seen in Table 4. Of all the types of food from table 1, there are only 6 out of 10 types of foods that contain omega-3. The highest amount of food consumed by all respondents was chicken eggs with an average of 35.07 mg/day. However, the average amount of food containing the highest omega-3 was chicken meat at 134.25 mg/day (42 of 49) (Table 4).

Table 4: Profiles of Types of Foods Containing Omega-3 per Day

The type of food consumed	Frequency (n:49)	Mean (mg)	SD	Minimum (mg)	Maximum (mg)
Chicken eggs	49	35.07	16.63	19.34	50.00
Chicken meat	42	134.25	24.66	9.72	201.57
Catfish	25	45.33	2.59	28.28	74.05
Shrimp	21	10.08	42.69	2.04	89.32
Beef	20	36.56	23.01	7.55	106.41
Sardines	15	47.5	3.41	15.73	76.63

4. DISCUSSION

Cigarettes had high concentration of free radicals and oxidants that induce systemic oxidative stress and lipid peroxidation thereby affecting levels of omega-3 PUFAs, particularly EPA and DHA [16]. Smoking increases the bioavailability of long-chain fatty acids from plasma and results in decreased serum omega-3 fatty acids [18]. The brain was susceptible to oxidative stress due to high metabolic activity and susceptibility of PUFAs to free radical attack, so smoking can also reduce levels of omega-3 PUFAs in brain tissue [41]. The long-chain omega-3 PUFAs (EPA and DHA) are important components of cell membranes and as a major part of brain tissue that play a key role in brain function and nerve transmission. Deficiency of omega-3 PUFAs is associated with mesocorticolimbic dysfunction in the dopamine pathway which triggers the release of dopamine, leading to nicotine addiction [42].

Foods that are widely consumed by respondents include:

a. Egg. Omega-3 fatty acids are important molecules because the human body cannot produce them, so the source of omega-3 fatty acids can be obtained from food [25- 27]. Eggs were the food that contains the most omega-3s consumed by respondents with an average consumption of 19.46 mg per day for 49 respondents. The total content of omega in ordinary eggs is 22 mg per 1 egg [28], [36], [37]. Whereas in eggs that are enriched with DHA, there are 100-150 mg of total omega-3 fatty acids in every 1 egg [43]. Eggs were naturally not rich in omega-3 fatty acids, therefore supplementation of omega-3 fatty acids in chicken feed is needed to get omega-3 fortified eggs. The omega-3 fatty acid content in eggs can be increased by supplementing the laying hens' diet with certain dietary supplements such as peanut oil, fish oil, safflower oil, flaxseed, fish meal, and microalgae [44]. Omega-3 fatty acids function as good fats for human health, therefore the content of omega-3 fatty acids in egg yolks helps reduce bad cholesterol content. The stability of omega-3 fatty acids can be increased with vitamin E and or organic selenium which reduces oxidation in raw eggs, thereby providing a protective effect during egg marketing, storage and cooking [44- 46]. However, the price of eggs enriched with DHA is too high, so that ordinary eggs tend to be chosen more for daily consumption. Eggs can be an alternative source of foods that contain omega-3s, as they are a very useful source for someone who is vegetarian and for people who do not eat fish regularly.

b. Chicken meat. The second most common food consumed by respondents is chicken with an average consumption of 10.74 mg per day for 47 respondents. There are 19 mg/100 g edible portion omega-3 in chicken [28]. In the chicken breast that is cooked in a way roasted each 85 grams there are 30 mg of total omega-3 [29]. Supplementation of omega-3 fatty acids in poultry feed especially in the form of EPA and DHA can improve the quality of meat. However, Omega-3 fatty acids are particularly susceptible to oxidative damage, which they produce bad taste and fishy smell so it will adversely affect consumer acceptance [47]. According to one of the motorcycle taxi drivers online when interviewed, chicken consumption has become a food habit because besides the price and the taste is good, chicken meat is available in various kinds processed. The second most common food consumed by respondents was chicken with an average consumption of 10.74 mg per day for 47 respondents. There are 19 mg/100 g edible portion

omega-3 in chicken meat. In the chicken breast that is roasted by means of 85 grams, there are 30 mg of total omega-3 [28], [44]. According to one online motorcycle taxi driver when being interviewed, chicken consumption has become a food habit because in addition to the price and good taste, chicken meat is available in a variety of preparations.

c. Catfish. The third food with an average consumption of 16.36 mg per day by 33 respondents is catfish. For every 85 grams of catfish raised there are 151 mg of total omega-3. Whereas for every 85 grams of wild catfish there are 201 mg of total omega-3 [40]. Data from National Nutrient Data by United States Department of Agriculture showed Greenland halibut, farmed catfish, wild catfish, farmed salmon, and wild salmon contained EPA+DHA at 1177.6, 177.6, 236.5, 2147.1, and 1840 mg/100 g muscles [20]. Generally, most samples in the current study contained EPA + DHA amounts (11.8–551.7 mg/100 g wet sample); which were lower compared to Greenland halibut, farmed salmon, and wild salmon; but comparable with farmed and wild catfish [48]. Catfish can be an alternative source of foods that contain omega-3 because of the price which is relatively cheap and the availability of catfish is quite high and easy to get.

d. Shrimp. Northern shrimp (*Pandalus borealis*) oil, which was rich in omega-3 fatty acids, was recovered from the cooking water of shrimp processing facilities. The oil contains significant amounts of omega-3 fatty acids in triglyceride form along with substantial long-chain monounsaturated fatty acids (MUFAs). Northern shrimp oil with PUFAs preferentially attached to the sn-2 positions of the TAG may present higher stability resulting in a favorable absorption of omega-3 fatty acids relative to other forms, which would be of benefit for use as a health supplement ingredient [49]. If consumed within reasonable limits, shrimp give health benefits, but if consumed too much, shrimp can cause various health problems such as an increase in cholesterol levels in blood. Shrimp contains 124 mg cholesterol and 0 g saturated fatty acids [50].

e. Beef. Beef contains omega-3 as much as 22 mg/100g [28]. The content of omega-3 found in beef was relatively low, and the price of beef is relatively expensive. Despite the content of saturated fatty acids in beef, it is also rich in heart healthy cis-monounsaturated fatty acids, and can be an important source of long-chain omega-3 fatty acids in populations where little or no oily fish is consumed. Beef also contains polyunsaturated fatty acid biohydrogenation products, including vaccenic and rumenic acids, which have been shown to have anticarcinogenic and hypolipidemic properties in cell culture and animal models. Beef can be enriched with these beneficial fatty acids through manipulation of beef cattle diets, which is now more important than ever because of increasing public understanding of the relationships between diet and health. The present review examines recommendations for beef in human diets, the need to recognize the complex nature of beef fat, how cattle diets and management can alter the fatty acid composition of beef, and to what extent content claims are currently possible for beef fatty acids [51].

f. Lamb. Lamb received an average of 0.6 mg per day for 14 respondents. Goat meat contains 18 mg of omega-3/100 g [28]. Lamb cannot be used as a food choice to meet the needs of omega-3 intake per day because low levels. The ratio of EPA+DHA from lambs with high values (97.5% quantile) to lambs with low values (2.5% quantile) also differed dramatically between locations, and between slaughters from the same location. Consistency between years, at a location, was less for the high to low value ratio of EPA plus DHA than for the median value of EPA plus DHA. To consistently obtain high levels of omega-3 fatty acids in Australian lamb, there must be a focus on lamb finishing diets which are likely to need a supply of α -linolenic acid (18:3n-3), the precursor for EPA+DHA [52].

The highest omega-3 content was found in salmon, herring and tuna. In 85 grams of farmed salmon there

are 1825 mg of total omega-3, herring contains 1807 mg of total omega-3 in 85 grams of serving, and tuna has omega-3 content of 1279 mg per 85 grams of serving. It can be seen that salted fish will be higher in omega-3 content compared to freshwater fish. Omega-3 in fish is also influenced by fish food intake. Usually fish that are farmed and which are more awake for food will also have a high omega-3 content. The state of fresh cooked fish compared to canned fish is also different. Research conducted by Stephen et.al. [53] shows that canning destroys omega-3 fatty acids. The cooking process of tuna does not cause major damage to the omega-3 fatty acids (EPA+ DHA), microwave heating causes a loss of 25% EPA and 55% DHA, while frying results in about 70% loss of EPA and 85% loss of DHA. This study shows that microwave cooking can be used to process tuna to retain omega-3 fatty acids. Different fish processing methods have different effects on the chemical, physical and nutritional composition of fish. This effect can affect the reduction of polyunsaturated fatty acids. The heat intensity given during processing greatly affects the concentration of fish nutrients, so it is necessary to know how temperature and processing time affect the nutritional properties and physico-chemical composition of fish and processed fish products [54].

The results of this study do not know what kind of processed process the respondents used, because at the time of the interview the researcher only asked about the frequency of meals and portion sizes. Salmon, herring and tuna may indeed be accessible to Indonesians. However, its high price reduces the ability of the Indonesian people to access it. Especially for online motorcycle taxi drivers, when interviewed they said they rarely even almost never eat sea fish. They tend to eat freshwater fish more often. Due to the price reason that makes it difficult for them to access it, coupled with the current pandemic condition, their decreasing average income has further hampered their intake of omega-3 foods which are computed from marine fish. Even salmon, tuna and herring were not included in the FFQ because none of the non-respondent respondents ate the fish.

Based on the results of research that has been done regarding omega-3 intake with the FFQ on online motorcycle taxi drivers who are active smokers can be given advice as following online motorcycle taxi drivers are advised to increase their intake consumption foods that contain omega-3 which can be obtained from foods that are affordable fish such as sardines, catfish, anchovies and other so that daily intake omega-3 is sufficient. It is necessary to do further research on the influencing factors lack of omega-3 intake in online motorcycle taxi drivers.

5. CONCLUSION

Based on the results of research conducted on 49 online motorcycle taxi drivers in the Rungkut area of Surabaya City, it can be concluded that online motorcycle taxi drivers are not consuming enough food that contains omega-3s. This can be seen from the average daily intake of 49 respondents who obtained 226.47 mg, where the average did not meet the daily recommendation of 1600 mg. The most consumed omega-3s by respondents were eggs and chicken, both of which contain low omega-3s.

6. ACKNOWLEDGMENTS

This research was funded by the Institute of Research and Community Service of the Universitas Surabaya.

7. CONFLICT OF INTEREST

The authors have no conflicts of interest regarding this investigation.

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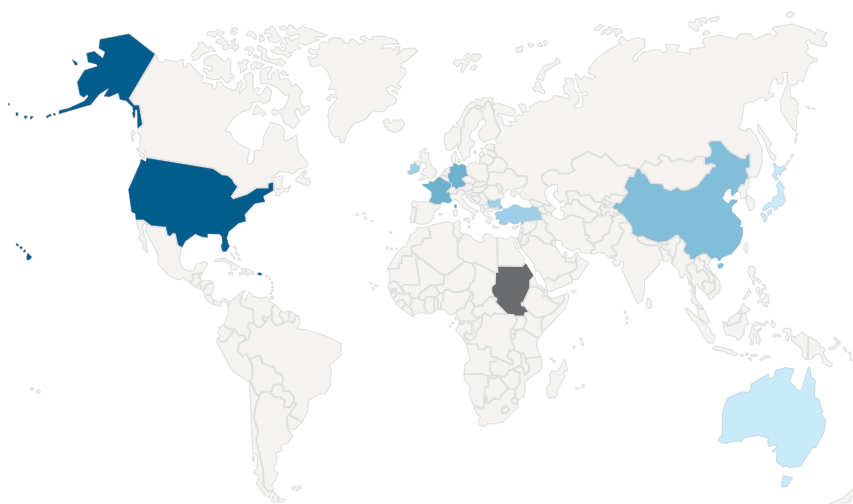
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Abstract :

Online motorcycle taxi drivers have risk of being exposed to vehicle air pollution, especially with smoking habits. Cigarette smoke can increase lipid peroxidation from PUFA (Polyunsaturated Fatty Acid) by triggering oxidative stress. Low concentrations of omega-3 fatty acids cause dysfunction in dopaminergic system associated with smoking dependence and craving. The purpose was to assess omega-3 fatty acid food intakes in online motorcycle taxi drivers. The research design was case-control method. Measurement of omega-3 intake was using FFQ and 24-hour recall. This research was conducted in Kali Rungkut, Surabaya City, in May-August 2020. The data analysis method used in this research was descriptive analysis. This research involved 49 respondents in the Rungkut area of Surabaya City, it can be concluded that they were not consuming enough food that contains omega-3. The results of omega-3 intake showed that all respondents were in the category of less intake of foods containing omega-3 (<1,600 mg) and average total intake of foods containing omega-3 was 226.47 mg. The most consumed omega-3s by respondents were eggs (an average of 19.46 mg/day) and chicken (an average of 10.74 mg/day), both of which contain low omega-3. Meanwhile, based on data collection per day with a 24-hour dietary recall, it was known that the average daily consumption of respondents is only 249.05 mg. Therefore, it was necessary to further analyze the processing pattern of the food consumed and the factors that influence their diet.

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Submission date: 30-Aug-2021 07:29AM (UTC+0700)

Submission ID: 1637860691

File name: food-intakes-in-online-motorcycle-taxi-drivers-611e1ca163244.pdf (254.03K)

Word count: 6303

Character count: 32560

Assessment of Omega-3 Fatty Acid Food Intakes in Online Motorcycle Taxi Drivers

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Keywords:

Active smokers, food Frequency Questionnaire, omega-3, online motorcycle taxi drivers, 24- hour recall.

ABSTRACT

Online motorcycle taxi drivers have risk of being exposed to vehicle air pollution, especially with smoking habits. Cigarette smoke can increase lipid peroxidation from PUFA (Polyunsaturated Fatty Acid) by triggering oxidative stress. Low concentrations of omega-3 fatty acids cause dysfunction in dopaminergic system associated with smoking dependence and craving. The purpose was to assess omega-3 fatty acid food intakes in online motorcycle taxi drivers. The research design was case-control method. Measurement of omega-3 intake was using FFQ and 24-hour recall. This research was conducted in Kali Rungkut, Surabaya City, in May-August 2020. The data analysis method used in this research was descriptive analysis. This research involved 49 respondents in the Rungkut area of Surabaya City, it can be concluded that they were not consuming enough food that contains omega-3. The results of omega-3 intake showed that all respondents were in the category of less intake of foods containing omega-3 (<1.600 mg) and average total intake of foods containing omega-3 was 226.47 mg. The most consumed omega-3s by respondents were eggs (an average of 19.46 mg/day) and chicken (an average of 10.74 mg/day), both of which contain low omega-3. Meanwhile, based on data collection per day with a 24-hour dietary recall, it was known that the average daily consumption of respondents is only 249.05 mg. Therefore, it was necessary to further analyze the processing pattern of the food consumed and the factors that influence their diet.



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1. INTRODUCTION

The main sources of air pollution come from various human activities such as industry, transportation, offices and housing. These various kinds of activities are the biggest contribution from air pollutants that discharged into the free air. The impact of air pollution can cause a decrease in air quality which will have a negative impact on human health [1]. In urban areas, especially in Surabaya, which is the largest metropolitan city in the province of East Java, the activities of its citizens really need fast access, especially

transportation. In today's modern era, people tend to choose fast and practical transportation, one of which is an online motorcycle taxi driver. This can have a high risk of decreased lung function, due to breathing air which is of bad quality for health from exhaust emissions from various motorized vehicles on the road [2], [3]. Decreased lung function can also be affected by smoking. Ninety percent of online motorcycle taxi drivers take advantage of their spare time while waiting for passengers by smoking cigarettes [4], [5]. Cigarettes consist of aerosols and gases, have thousands of chemical components and contain many toxins and are carcinogens. Some of the chemicals contained in cigarettes include nicotine, tar, carbon monoxide and various heavy metals [6], [7]. Nicotine is a drug in the form of tobacco that causes addiction, and nicotine dependence was an indirect root cause for decreased lung function of smokers and showed a gradual decrease in FEV1 with an increase in nicotine dependence [8], [9]. Cigarette smoke is a free radical obtained from exogenous sources. Cigarette smoke is also the main factor affecting the increase in free radicals in the body. An increase in the number of free radicals in the body will trigger oxidative stress and stimulate peroxidation in cells, which can cause damage and death to body cells [10], [11]. Cigarette smoke as free radicals increases Reactive Oxygen Species (ROS) which will result in oxidative stress in the lungs. An imbalance of oxidants and antioxidants in the body causes oxidative stress and triggers an inflammatory reaction in the alveolar tissue. The inflammatory process results in active lung macrophages and neutrophil infiltration which then causes inactivation of the α -AT1 antiprotease, as a pulmonary protease inhibitor in producing pulmonary elastase [12- 14]. This can damage the protein structure of the lung, through the destruction of the alveolar septum so that the elasticity of the lung parenchyma tissue is disrupted and causes changes in the structure and function of the lung tissue [14], [15].

Cigarette smoke can increase lipid peroxidation from PUFA (Polyunsaturated Fatty Acid) by triggering oxidative stress. Low concentrations of PUFA will affect neurotransmission in the central nervous system, namely dopaminergics, which will increase nicotine addiction, thereby inhibiting smoking cessation efforts [16], [17]. Previous research conducted by [18] showed that smokers consume less fish rich in omega-3 fatty acids compared to nonsmokers according to lower levels of DHA (docosahexaenoic acid) and EPA (eicosapentaenoic Acid). The importance of omega-3 intake in smoking, because it can normalize the dopaminergic system so as to reduce addiction. Omega-3 fatty acids were effective in significantly reducing smoking cravings. Cigarettes have a high concentration of free radicals and oxidants that induce systemic oxidative stress and lipid peroxidation, thereby affecting levels of omega-3 PUFAs, particularly EPA and DHA. Smoking increases the bioavailability of long-chain fatty acids from plasma and results in decreased levels of serum omega-3 fatty acids. The brain is susceptible to oxidative stress due to high metabolic activity and susceptibility of PUFAs to free radical attack, so smoking can also reduce levels of omega-3 PUFAs in brain tissue. The long-chain omega-3 PUFAs (EPA and DHA) are important components of cell membranes and is a major part of brain tissue that play a key role in brain function and nerve transmission. Deficiency of omega-3 PUFAs is associated with mesocorticolimbic dysfunction in the dopamine pathway which triggers the release of dopamine, leading to nicotine addiction [16- 19].

Omega-3s have beneficial effects on chronic inflammatory diseases including chronic obstructive pulmonary disease (COPD), asthma, rheumatoid arthritis, and inflammatory bowel disease [20]. Higher omega-3 intake and fish consumption also predict better lung function in smokers and ex-smokers with EPA and DHA acting as antioxidants and reducing oxidative stress [18], [21]. Epidemiological studies show EPA and DHA are the main omega-3 PUFAs found in fish, fish oil and fish oil supplements [22- 24]. In addition, omega-3s can be obtained easily from foods such as salmon, sardines, seafood, and others [25]. According to Recommended Nutritional Adequacy Rate for Indonesian People, concerning the recommended nutritional adequacy rate, for omega-3s of 1.6 g/day in men ≥ 18 years [26]. The pattern of omega-3 intake can be determined by using the FFQ (Food Frequency Questionnaire). The FFQ is a method

of assessing food consumption. The FFQ method is a semi-qualitative method, in which information about the food ingredients consumed is only in the form of names, while the amounts are not clearly differentiated. Every subject who states that they often consume certain foods and drinks, does not always have to be further broken down into the size and portion consumed. The FFQ method only requires data that certain types of food are often or not frequently consumed and their frequency [27- 31]. Previous research conducted by [31] showed that the FFQ can be used to categorize omega-3 fatty acid status. The data collection omega-3 intake in a day is reinforced with a 24-hour recall method, a structured interview intended to capture detailed information about all foods and beverages containing omega-3 [28], [29]. The type of food consumed by a person is strongly influenced by a person's socioeconomic status, cause lower food expenditure is likely to be a key contributor to less-healthy food choices [31]. The purpose of this study was to assess omega-3 fatty acid food Intakes in online motorcycle taxi drivers.

2. METHODS

2.1 Design

The research design used in this study was an observational study, with a case-control method. Measurement of omega-3 intake was carried out using the FFQ and 24-hour recall. This research will be conducted in May- August 2020. This research was conducted in Kali Rungkut, Surabaya City. This research had received ethical permission from the University of Surabaya Ethics Committee Number 016-OL/KE.VII/2020.

2.2 Research Variabel

The variable in this study was the intake of foods containing omega-3. Online motorcycle taxi drivers were public transportation drivers in Indonesia in the form of motorbikes rented by passengers. Orders and payments can be made via the application. An active smoker was someone who has smoked more than 100 cigarettes their lifetime, and was still consuming cigarettes in the past 28 days. A non-smoker was someone who has never smoked or smoked less than 100 cigarettes in his lifetime [32].

Omega-3 intake was the intake consumed by respondents daily using the FFQ. Researchers will interview respondents with the FFQ which contains the frequency of meals, the number of times the food has been consumed per day, week, month or never. Regular consumption of fish will protect the lungs of smokers by targeting a daily intake of about 1 gram per day of fish or supplements [33]. Many sources of omega-3 were found in salmon, seafood, tuna, fish oil supplements, eggs, soybean oil, canola oil, and others [34], [35]. The way to calculate the omega-3 content was to convert the portion size then look at the list of foods that contain omega-3, then calculate the omega-3 content in food multiplied by the frequency of food from the respondent. The calculation result of the omega-3 content of all foods containing omega-3 in units of milligrams. After that it was compared with the nutritional adequacy rate (RDA) of omega-3 in Indonesia, where if 1600 mg is categorized as sufficient, if <1600 mg is categorized as insufficient [27].

2.3 Population and Sample Research

The population in this study were online motorcycle taxi drivers operating in the Kali Rungkut area of Surabaya City. The sample in this study were online motorcycle taxi drivers operating in the Kali Rungkut area of Surabaya City who met the following criteria: (1) Male; (2) ≥ 18 years old; (3) Active smokers (smoke ≥ 30 cigarette/day); (5) Working 6-8 hours/day; (6) Didn't not have chronic lung function disorders such as asthma, COPD (chronic obstructive pulmonary disease), and respiratory infections; and (7) Not eating vegetarian or being on a certain diet. The sampling technique used purposive sampling and snowball sampling.

2.4 Method of collecting data in Statistics & Data analysis method

The instrument used in this study used a FFQ consisting of 2 parts, namely: demographic data and consumption patterns of food intake containing omega-3 (shrimp, fish sardines, anchovies, catfish, fish salmon, tuna, lobster, crab, shells, eggs, meat chicken, beef, lamb) (Table 1). Preparation for making a questionnaire with several steps as follows: (1) Find list of foods containing omega-3 based on foods that were often consumed by Indonesians [28], [36], [37].

Table 1: List of Composition of Food Intake Containing Omega-3 [28], [36], [37]

The type of food consumed	gram	Omega-3 content (mg/serving)
Eggs	63	22
Chicken meat	100	19
Catfish	85	151
Shrimp	85	267
Beef	100	22
Sardines	57	556
Lamb	100	18
Anchovies	57	1,165
Shellfish	85	241
Crab	85	351
Lobster	100	84
Salmon	85	1,825
Tuna	85	228

The frequency of eating can be calculated in terms of the number of times the food has been consumed each day, week, month or never. This was the first time conducting a preliminary study on online motorcycle taxi drivers who are not research respondents to find out the food on the list that has been made available or often consumed in the area around the research location. This study was conducted by interviewing 20 respondents. Foodstuffs that were consumed by less than 10% of the respondents because they have never been consumed or are not usually consumed are excluded from the food list. Then carry out the final stages or finalization for a list of foods that will be used in the FFQ [29], [30], [38]. After carrying out the final stages or finalizing the FFQ, data collection can be carried out on respondents who had met the criteria. From the interview data, it was processed by estimating the portion size of the food consumed by the respondent into weight (grams) to be able to get the weight of the food consumed in grams/day. After counting all the food items consumed, the known weight consumed in a day (grams/day) was recalculated by looking at the list of foods containing omega-3 and then multiplying by the frequency of meals of the respondent. The data analysis method used in this research was descriptive analysis. Descriptive analysis was method used to describe or describe the collected data without intending to make general conclusions or generalizations.

3. RESULTS

The research design used in this study was an observational study, with case-control method. Measurement of omega-3 intake was carried out using FFQ and 24-hour dietary recall. At the location there were 60 respondents of online motorcycle taxi drivers who met the criteria and only 49 respondents were willing to take part in this study. The rest, as many as 11 respondents, online motorcycle taxi drivers had several reasons so that they were not willing to take part in this research.

The average age being 35.10 years. The lowest age was 21 years, while the highest age was 56 years. Of the 49 respondents, 14 of 49 consumed 1-10 cigarettes per day, 32 of 49 consumed 11-20 cigarettes per day, and 3 of 49 consumed >20 cigarettes per day. Most of the respondents included based on the Brinkman

index were included in mild smokers level (32 of 49). In addition, education level most of the respondents are senior high school (36 of 49) (Table 2).

Table 2: Frequency Distribution of Respondents

Characteristics	Frequency (n=49)	Percentage (%)	
Age (years)	18-25	9	18.40
	26-35	17	34.70
	36-45	17	34.70
	46-55	4	8.20
	56-65	2	4.10
Number of Cigarette per day [39]	1-10	14	28.57
	11-20	32	65.31
	>20	3	6.12
Duration of smoking [39]	<10	17	34.69
	10-20	23	46.94
	<20	9	18.37
Brinkman Index [40]	Mild (0-199)	32	65.3
	Moderate (200-600)	15	32.65
	Severe (>600)	2	4.08
Level of education	Elementary school	3	6.10
	Junior high school	2	4.10
	Senior high school	36	73.50
	Bachelor degree	8	16.30

The results of omega-3 intake showed that all respondents were in the category of less intake of foods containing omega-3 (<1,600 mg). The average total intake of foods containing omega-3 was 226.47 mg, with minimum value was 9.43 mg and maximum value was 1,116.34 mg. The type of food containing omega-3 that was consumed the most was chicken eggs by all respondents (average: 19.46 mg/day), although when calculated from the omega-3 content in eggs only 22 mg/serving of 63 g eggs. The least type of food was crab in 5 of the 49 respondents (average: 16.38 mg/day) (Table 3).

Table 3: Profile Distribution of Foods Containing Omega-3 by FFQ

The type of food consumed	Frequency (n=49)	Mean (mg)	SD	Minimum (mg)	Maximum (mg)
Chicken eggs	49	19.46	15.41	0.73	66.00
Chicken meat	47	10.74	4.76	0.63	30.40
Catfish	33	16.36	16.31	2.37	71.05
Shrimp	30	107.04	142.19	8.90	801.00
Beef	26	3.53	4.32	0.73	12.57
Sardines	17	192.19	224.64	18.53	556.00
Lamb	14	0.60	1.20	0.60	0.60
Anchovies	12	156.51	221.92	9.43	613.16
Shellfish	10	55.66	50.21	8.03	103.29
Crab	5	16.38	10.46	11.7	35.1

Meanwhile, based on data collection per day with a 24-hour dietary recall, it was known that the average daily consumption of respondents is only 249.05 mg, with the minimum value being 0 mg, the maximum value was 1069.43 mg. The profile of food consumed per day containing omega-3 can be seen in Table 4. Of all the types of food from table 1, there are only 6 out of 10 types of foods that contain omega-3. The highest amount of food consumed by all respondents was chicken eggs with an average of 35.07 mg/day. However, the average amount of food containing the highest omega-3 was chicken meat at 134.25 mg/day (42 of 49) (Table 4).

Table 4: Profiles of Types of Foods Containing Omega-3 per Day

The type of food consumed	Frequency (n:49)	Mean (mg)	SD	Minimum (mg)	Maximum (mg)
Chicken eggs	49	35.07	16.63	19.34	50.00
Chicken meat	42	134.25	24.66	9.72	201.57
Catfish	25	45.33	2.59	28.28	74.05
Shrimp	21	10.08	42.69	2.04	89.32
Beef	20	36.56	23.01	7.55	106.41
Sardines	15	47.5	3.41	15.73	76.63

4. DISCUSSION

Cigarettes had high concentration of free radicals and oxidants that induce systemic oxidative stress and lipid peroxidation thereby affecting levels of omega-3 PUFAs, particularly EPA and DHA [16]. Smoking increases the bioavailability of long-chain fatty acids from plasma and results in decreased serum omega-3 fatty acids [18]. The brain was susceptible to oxidative stress due to high metabolic activity and susceptibility of PUFA to free radical attack, so smoking can also reduce levels of omega-3 PUFAs in brain tissue [41]. The long-chain omega-3 PUFAs (EPA and DHA) are important components of cell membranes and a major part of brain tissue that play a key role in brain function and nerve transmission. Deficiency of omega-3 PUFAs is associated with mesocorticolimbic dysfunction in the dopamine pathway which triggers the release of dopamine, leading to nicotine addiction [42].

Foods that are widely consumed by respondents include:

a. Egg. Omega-3 fatty acids are important molecules because the human body cannot produce them, so the source of omega-3 fatty acids can be obtained from food [25- 27]. Eggs were the food that contains the most omega-3s consumed by respondents with an average consumption of 19.46 mg per day for 49 respondents. The total content of omega in ordinary eggs is 22 mg per 1 egg [28], [36], [37]. Whereas in eggs that are enriched with DHA there are 100-150 mg of total omega-3 fatty acids in every 1 egg [43]. Eggs were naturally not rich in omega-3 fatty acids, therefore supplementation of omega-3 fatty acids in chicken feed is needed to get omega-3 fortified eggs. The omega-3 fatty acid content in eggs can be increased by supplementing the laying hens' diet with certain dietary supplements such as peanut oil, fish oil, safflower oil, flaxseed, fish meal, and microalgae [44]. Omega-3 fatty acids function as good fats for human health, therefore the content of omega-3 fatty acids in egg yolks helps reduce bad cholesterol content. The stability of omega-3 fatty acids can be increased with vitamin E and or organic selenium which reduces oxidation in raw eggs, thereby providing a protective effect during egg marketing, storage and cooking [44- 46]. However, the price of eggs enriched with DHA is too high, so that ordinary eggs tend to be chosen more for daily consumption. Eggs can be an alternative source of foods that contain omega-3s, as they are a very useful source for someone who is vegetarian and for people who do not eat fish regularly.

b. Chicken meat. The second most common food consumed by respondents is chicken with an average consumption of 10.74 mg per day for 47 respondents. There are 19 mg/100 g edible portion omega-3 in chicken [28]. In the chicken breast it is cooked in a way roasted each 85 grams there are 30 mg of total omega-3 [29]. Supplementation of omega-3 fatty acids in poultry feed especially in the form of EPA and DHA can improve the quality of meat. However, Omega-3 fatty acids are particularly susceptible to oxidative damage, which they produce bad taste and fishy smell so it will adversely affect consumer acceptance [47]. According to one of the motorcycle taxi drivers online when interviewed, chicken consumption has become a food habit because besides the price and the taste is good, chicken meat is available in various kinds processed. The second most common food consumed by respondents was chicken with an average consumption of 10.74 mg per day for 47 respondents. There are 19 mg/100 g edible portion

omega-3 in chicken meat. In the chicken breast that is roasted by means of 85 grams, there are 30 mg of total omega-3 [28], [44]. According to one online motorcycle taxi driver when being interviewed, chicken consumption has become a food habit because in addition to the price and good taste, chicken meat is available in a variety of preparations.

c. Catfish. The third food with an average consumption of 16.36 mg per day by 33 respondents is catfish. For every 85 grams of catfish raised there are 151 mg of total omega-3. Whereas for every 85 grams of wild catfish there are 201 mg of total omega-3 [40]. Data from National Nutrient Data by United States Department of Agriculture showed Greenland halibut, farmed catfish, wild catfish, farmed salmon, and wild salmon contained EPA+DHA at 1177.6, 177.6, 236.5, 2147.1, and 1840 mg/100 g muscles [20]. Generally, most samples in the current study contained EPA + DHA amounts (11.8–551.7 mg/100 g wet sample); which were lower compared to Greenland halibut, farmed salmon, and wild salmon; but comparable with farmed and wild catfish [48]. Catfish can be an alternative source of foods that contain omega-3 because of the price which is relatively cheap and the availability of catfish is quite high and easy to get.

d. Shrimp. Northern shrimp (*Pandalus borealis*) oil, which was rich in omega-3 fatty acids, was recovered from the cooking water of shrimp processing facilities. The oil contains significant amounts of omega-3 fatty acids in triglyceride form along with substantial long-chain monounsaturated fatty acids (MUFAs). Northern shrimp oil with PUFAs preferentially attached to the sn-2 positions of the TAG may present higher stability resulting in a favorable absorption of omega-3 fatty acids relative to other forms, which would be of benefit for use as a health supplement ingredient [49]. If consumed within reasonable limits, shrimp give health benefits, but if consumed too much, shrimp can cause various health problems such as an increase in cholesterol levels in blood. Shrimp contains 124 mg cholesterol and 0 g saturated fatty acids [50].

e. Beef. Beef contains omega-3 as much as 22 mg/100g [28]. The content of omega-3 found in beef was relatively low and the price of beef is relatively expensive. Despite the content of saturated fatty acids in beef, it is also rich in heart healthy cis-monounsaturated fatty acids, and can be an important source of long-chain omega-3 fatty acids in populations where little or no oily fish is consumed. Beef also contains polyunsaturated fatty acid biohydrogenation products, including vaccenic and rumenic acids, which have been shown to have anticarcinogenic and hypolipidemic properties in cell culture and animal models. Beef can be enriched with these beneficial fatty acids through manipulation of beef cattle diets, which is now more important than ever because of increasing public understanding of the relationships between diet and health. The present review examines recommendations for beef in human diets, the need to recognize the complex nature of beef fat, how cattle diets and management can alter the fatty acid composition of beef, and to what extent content claims are currently possible for beef fatty acids [51].

f. Lamb. Lamb received an average of 0.6 mg per day for 14 respondents. Goat meat contains 18 mg of omega-3/100 g [28]. Lamb cannot be used as a food choice to meet the needs of omega-3 intake per day because low levels. The ratio of EPA+DHA from lambs with high values (97.5% quantile) to lambs with low values (2.5% quantile) also differed dramatically between locations, and between slaughters from the same location. Consistency between years, at a location, was less for the high low value ratio of EPA plus DHA than for the median value of EPA plus DHA. To consistently obtain high levels of omega-3 fatty acids in Australian lamb, there must be a focus on lamb finishing diets which are likely to need a supply of α -linolenic acid (18:3n-3), the precursor for EPA+DHA [52].

The highest omega-3 content was found in salmon, herring and tuna. In 85 grams of farmed salmon there

are 1825 mg of total omega-3, herring contains 1807 mg of total omega-3 in 85 grams of serving, and tuna has omega-3 content of 1279 mg per 85 grams of serving. It can be seen that salted fish will be higher in omega-3 content compared to freshwater fish. Omega-3 in fish is also influenced by fish food intake. Usually fish that are farmed and which are more awake for food will also have a high omega-3 content. The state of fresh cooked fish compared to canned fish is also different. Research conducted by Stephen et.al. [53] shows that canning destroys omega-3 fatty acids. The cooking process of tuna does not cause major damage to the omega-3 fatty acids (EPA+ DHA), microwave heating causes a loss of 25% EPA and 55% DHA, while frying results in about 70% loss of EPA and 85% loss of DHA. This study shows that microwave cooking can be used to process tuna to retain omega-3 fatty acids. Different fish processing methods have different effects on the chemical, physical and nutritional composition of fish. This effect can affect the reduction of polyunsaturated fatty acids. The heat intensity given during processing greatly affects the concentration of fish nutrients, so it is necessary to know how temperature and processing time affect the nutritional properties and physico-chemical composition of fish and processed fish products [54].

The results of this study do not know what kind of processed process the respondents used, because at the time of the interview the researcher only asked about the frequency of meals and portion sizes. Salmon, herring and tuna may indeed be accessible to Indonesians. However, its high price reduces the ability of the Indonesian people to access it. Especially for online motorcycle taxi drivers, when interviewed they said they rarely even almost never eat sea fish. They tend to eat freshwater fish more often. Due to the price reason that makes it difficult for them to access it, coupled with the current pandemic condition, their decreasing average income has further hampered their intake of omega-3 foods which are computed from marine fish. Even salmon, tuna and herring were not included in the FFQ because none of the non-respondent respondents ate the fish.

Based on the results of research that has been done regarding omega-3 intake with the FFQ on online motorcycle taxi drivers who are active smokers can be given advice as following online motorcycle taxi drivers are advised to increase their intake consumption foods that contain omega-3 which can be obtained from foods that are affordable fish such as sardines, catfish, anchovies and other so that daily intake omega-3 is sufficient. It is necessary to do further research on the influencing factors lack of omega-3 intake in online motorcycle taxi drivers.

5. CONCLUSION

Based on the results of research conducted on 49 online motorcycle taxi drivers in the Rungkut area of Surabaya City, it can be concluded that online motorcycle taxi drivers are not consuming enough food that contains omega-3s. This can be seen from the average daily intake of 49 respondents who obtained 226.47 mg, where the average did not meet the daily recommendation of 1600 mg. The most consumed omega-3s by respondents were eggs and chicken, both of which contain low omega-3s.

6. ACKNOWLEDGMENTS

This research was funded by the Institute of Research and Community Service of the Universitas Surabaya.

7. CONFLICT OF INTEREST

The authors have no conflicts of interest regarding this investigation.

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