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eJournal Kedokteran Indonesia (eJKE) is a general medical journal, published quarterly (April, August, December) by Faculty of Medicine Universitas Indonesia, Jakarta, Indonesia. The official journal title is eJournal Kedokteran Indonesia and the abbreviated title is eJKE.

Authors can submit manuscripts to eJKE through the website. All correspondences to the corresponding author will be sent electronically.

Last accredited (2019-2023) by Directorate General of Higher Education, Research, and Technology of the Ministry of Education, Culture, Research, and Technology of the Republic of Indonesia with Second Grade (<https://sinta.kemdikbud.go.id/journals/detail?page=15&id=1662>)

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PUBLISHED: 2023-01-10

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High Urinary Cotinine Levels and Low Academic Performance of Elementary School Students in Families with Careless Smoking Habits

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Keywords: Smoking habits, Urinary cotinine, Passive smoking, SHS, THS

ABSTRACT

Children are the most vulnerable age group to become passive smokers due to cigarette smoke exposure, especially from family members. In addition, children who smoke passively are at high risk of experiencing declined cognitive performance. This study investigated the effect of cigarette smoke exposure on urinary cotinine levels and its relationship with elementary school students' academic performance. The observational study with a cross-sectional design was located in Pannampu Village, Tallo District, Kota Makassar. Interviews, collecting academic performance data, and measuring urinary cotinine levels using colorimetric techniques were carried out on elementary school students (n=77). This study found that 88.5% of students stated that they had family members who smoked at home with inappropriate habits of exposing children to cigarette smoke. Urinary cotinine levels showed an average of 121.64 ng/mL. All students with urinary cotinine levels above 200 ng/mL (16.4%) had smoker family members. The urinary cotinine levels in boys were higher ($p=0.014$) with lower report card average marks than in girls. The smoking habits of family members were still a concern and had not protected children from cigarette smoke exposure, as evidenced by the high average urinary cotinine levels, especially in boys, which might affect their academic performance

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eJKI
eJournal Kedokteran Indonesia
ejki.fk.ui.ac.id

Volume 10, Nomor 3
Desember 2022

ISSN Print:2338-1426
ISSN Online:2338-6037

eJournal Kedokteran Indonesia

Editorial :

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FAKULTAS
KEDOKTERAN

PUBLISHED

2023-01-10

HOW TO CITE

Yustisia, I., Sari, N., Tawali, S., Arif, M., & Ramliah, S. (2023). High Urinary Cotinine Levels and Low Academic Performance of Elementary School Students in Families with Careless Smoking Habits. *EJournal Kedokteran Indonesia*, 10(3). Retrieved from <https://ejki.fk.ui.ac.id/index.php/journal/article/view/199>

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Research Article

High Urinary Cotinine Levels and Low Academic Performance of Elementary School Students in Families with Careless Smoking Habits

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Received 6 April 2022; Accepted 8 December 2022

<https://doi.org/10.23886/ejki.10.199.197>

Abstract

Children are the most vulnerable age group to become passive smokers due to cigarette smoke exposure, especially from family members. In addition, children who smoke passively are at high risk of experiencing declined cognitive performance. This study investigated the effect of cigarette smoke exposure on urinary cotinine levels and its relationship with elementary school students' academic performance. The observational study with a cross-sectional design was located in Pannampu Village, Tallo District, Kota Makassar. Interviews, collecting academic performance data, and measuring urinary cotinine levels using colorimetric techniques were carried out on elementary school students ($n=77$). The research activities took place from June to August 2018. This study found that 88.5% of students stated that they had family members who smoked at home with inappropriate habits of exposing children to cigarette smoke. Urinary cotinine levels showed an average of 121.64 ng/mL. All students with urinary cotinine levels above 200 ng/mL (16.4%) had smoker family members. The urinary cotinine levels in boys were higher (Mann-Whitney test, $p=0.014$) with lower report card average marks than in girls. The smoking habits of family members were still a concern and had not protected children from cigarette smoke exposure, as evidenced by the high average urinary cotinine levels, especially in boys, which might affect their academic performance.

Keywords: smoking habits, urinary cotinine, passive smoking.

Kadar Kotinin Urin dan Prestasi Belajar Siswa Sekolah Dasar pada Keluarga dengan Perilaku Merokok yang Buruk

Abstrak

Anak-anak merupakan kelompok usia paling rentan menjadi perokok pasif akibat paparan asap rokok dari orang-orang sekeliling terutama anggota keluarga. Anak-anak perokok pasif berisiko tinggi mengalami gangguan kesehatan termasuk penurunan kemampuan kognitif. Penelitian ini bertujuan menganalisis pengaruh asap rokok terhadap kadar kotinin urin dan hubungannya dengan prestasi belajar anak. Penelitian observasional analitik dengan desain potong lintang ini berlokasi di Kelurahan Pannampu, Kecamatan Tallo, Kota Makassar. Wawancara, pengumpulan data prestasi akademik, dan pengukuran kadar kotinin urin dengan teknik kolorimetri dilakukan pada siswa usia sekolah dasar ($n=77$). Penelitian dilakukan pada bulan Juni-agustus 2018. Didapatkan 88,5% siswa memiliki anggota keluarga perokok di rumah dengan perilaku berisiko memajankan asap rokok pada anak. Kadar kotinin urin menunjukkan nilai rerata 121,64 ng/mL. Semua siswa dengan kadar kotinin urin >200 ng/mL (16,4%) memiliki anggota keluarga perokok di rumah. Kadar kotinin urin pada anak laki-laki lebih tinggi (Mann-Whitney test, $p=0,014$) dengan rerata nilai rapor lebih rendah dibandingkan anak perempuan. Perilaku merokok anggota keluarga memprihatinkan dan belum melindungi anak dari paparan asap rokok yang dibuktikan dengan rerata kadar kotinin urin yang tinggi terutama pada anak laki-laki. Diperlukan upaya yang kuat untuk melindungi anak dari bahaya rokok yang dapat bermanifestasi penurunan fungsi kognitif di masa depan.

Kata kunci: perilaku merokok, kotinin urin, perokok pasif.

Introduction

Cigarette smoking and other forms of tobacco consumption are public health problem that is part of behavioral health risk factors (HRFs).¹ Although many studies prove the health effects of cigarette smoking, education and campaigns are still being promoted; however, the proportion of smokers has not changed significantly. For example, data from Indonesian Basic Health Research (Riset Kesehatan Dasar/Riskesdas) showed that the proportion of smokers in Indonesia was 29.3% in 2013, only a slight decrease to 28.9% in 2018.^{2,3} In South Sulawesi Province, the proportion of smokers also decreased from 27.0% to 25.9%.⁴ However, these proportions are still high, considering that the global target for reducing smoking prevalence is 15.5% by 2025.¹

When a cigarette is burned, the smoke is formed, containing nicotine and other toxic chemical compounds that are harmful to health. Cigarette smoke inhaled by smokers is referred to as primary cigarette smoke (first-hand smoke = FHS).⁵ Meanwhile, smoke from the burning end of a cigarette and is exhaled by smokers is called second-hand smoke (SHS).^{5,6} The smoke produced from burning cigarettes does not evaporate into the air but still contains nicotine residue that can pollute the air. It can react with other chemicals to form carcinogens and then stick to the surface of surrounding objects such as clothes, carpets, walls, furniture, bed linen, car dashboards, and children's toys. This latter type of cigarette smoke, referred to as third-hand smoke (THS), can remain in the environment for years.^{5,6} Nicotine and tobacco-specific nitrosamines were common contaminants found on many indoor surfaces, including floors, walls, and windows.^{7,8} Nicotine derived from THS can enter the body through the skin, respiratory tract, and digestive tract.⁹

Children are the most vulnerable group to receive SHS and THS from smokers around them. The main source of cigarette smoke exposure to children is from their homes and surroundings.^{10,11} Individuals exposed to SHS, including children are susceptible to receiving more than 250 carcinogenic compounds and hazardous chemicals.¹² SHS exposure to children is likely to cause premature death and increase the risk of developing acute respiratory infections, severe asthma symptoms, ear infections, and cancers.^{1,10} In addition, SHS exposure and most likely THS can affect brain development because this organ is very sensitive to toxins.^{13,14} This study aims to evaluate the impact of SHS and THS exposure, especially the

nicotine component, on the academic performance of elementary school students. The intensity of nicotine exposure in children is determined by measuring the levels of the main metabolite of nicotine, namely cotinine, in urine.

Method

The research protocol has received ethical approval from the Ethics Commission of the Faculty of Medicine, Hasanuddin University (number 819/H.4.8.4.5.31/PP36-KOMETIK/ 2018). This analytic observational study with a cross-sectional design was conducted in Pannampu Village, Tallo District, Kota Makassar, centered on SD Inpres Pannampu III. The study began with education about the negative impact of smoking on children's health, then explained the research objectives and procedures, conducted informed consent, interviewed subjects, collected morning urine samples and academic performance data in the form of report cards. The activities took place from June to August 2018. The study population was elementary school students by taking a total sample of SD Inpres Pannampu III that met the inclusion criteria included 5th and 6th grade students, aged ten-year-old and over, received permission from their parents, proficient in reading and writing. Students were excluded if they decided to leave their willingness to be research subjects and were not cooperative during interviews and urine collection.

Interview

In the interview, subjects were asked to answer several questions including personal data (name, age, residential address, how many children are in the family, residential address, and class at school), family characteristics (number of family members, parents' occupations, number of family members outside the nuclear family, and number of families who smoke), and smoking habits and activities of family members.

Urine Collection and Cotinine Level Determination

Research subjects were asked to collect their morning urine into the pot after obtaining their parents' permission and signing the informed consent form. The urine samples were then picked up and transported to the Biochemistry Laboratory, Faculty of Medicine, Hasanuddin University using an icebox for cotinine levels measurement.

Cotinine levels in urine were measured using a colorimetric method modified from previous study.¹⁴

In summary, 0.5 mL of urine sample was added with 0.5 mL of ethanol, 0.2 mL of acetic buffer (4 M, pH 4.7), 0.1 mL of 1.5 M KCN, 0.1 mL of 0.44 M chloramine, and 0.5 mL of 78 mM barbituric acid. The solution mixture was incubated for 100 minutes at room temperature. The color formed from the solution was measured at a wavelength of 508 nm using a Genesys 150 UV VIS spectrophotometer (Thermo Scientific). Each sample was measured in triplicate. The absorbance value obtained was then converted to a concentration value based on the standard cotinine (Sigma C-016) curve, developed using the CurveExpert software.

Data Analysis

Data on academic performance used the average marks of report cards in the last two semesters. The data were collected and processed using Microsoft Excel 2016 software. Statistical analysis was carried out using the SPSS 20 for Windows.

Results

Out of 77 Grade 5 and Grade 6 students at SD Inpres Pannampu III who met the inclusion criteria, 61 students participated in the study until the end (Table 1). Sixteen students were uncooperative during the interview and did not collect urinary samples.

Smoking Habits of Family Members

The interview results showed several important points regarding the smoking habits of family members in the house. Of the 61 students interviewed, 54 students (88.5%) stated that they had at least one smoker family member at home and 7 students (11.5%) did not have smoker family members. From the students who had smoker family members, the smokers often smoked when

they were with the students (75.4%), when they gathered with the family (62.3%), while watching TV (68.9%) and after meals (52.4%). However, most students stated (73.8%) that other family members had advised smokers to stop or reduce cigarette consumption and not smoke inside the house.

Urinary Cotinine Level

The urinary cotinine levels (Table 1), both those who have family members who smoke at home and who did not have an average of 121.64 ng/mL ranging between 0.83 ng/mL to 655.78 ng/mL. This urinary cotinine level was not significantly different in the four age groups. Boys had significantly higher ($p=0.014$) level (173.20 ng/mL) than girls (74.90 ng/mL). Although not statistically significant, the greater the number of smoker family members and the higher the ratio of smokers per total number of family members at home, the higher the students' urinary cotinine level. Students whose fathers worked as traders in the traditional market or stalls, odd jobs, and laborers tended to have higher urinary cotinine levels. Students whose mothers work at home as homemakers have lower urinary cotinine levels than students whose mothers have certain professions such as traders/sellers in the traditional market or stalls, midwives, tailors, or small entrepreneurs.

The wide range of urinary cotinine levels (0.83 ng/mL – 655.78 ng/mL) was grouped into three as presented in Table 2. Most of the students ($n=39$; 63.93%) had urinary cotinine levels of 10 – 100 ng/mL and 87.2% of them had smoker family members. Students with cotinine concentrations \geq 200 ng/mL were a fairly high percentage (16.4%) and all of them had family members who smoked at home. So that there was a tendency for students who have smoker family members at home to have urinary cotinine levels higher than 10 ng/mL.

Table 1. Students' Urinary Cotinine Levels on Assessed Variables

Variables	n (%)	Urinary Cotinine Levels (ng/mL)#	p value
Ages (yo)			
10	21 (34.43)	132.32 ± 155.49	0.544
11	26 (42.62)	116.37 ± 142.54	
12	13 (21.31)	115.84 ± 59.22	
13	1 (1.64)	109.69	
Sex*			
Male	29 (47.54)	173.20 ± 171.81	0.014
Female	32 (52.46)	74.90 ± 45.35	
Number of family in the house			
4-6	28 (45.90)	141.81 ± 156.76	0.384
7-9	21 (34.43)	79.72 ± 47.64	
≥ 10	12 (19.67)	147.92 ± 157.18	
Number of smoker family member			
None	7 (11.48)	68.41 ± 55.74	0.688
1	27 (44.26)	130.28 ± 136.68	
2 - 4	20 (32.79)	133.69 ± 151.70	
≥ 5	7 (11.48)	107.09 ± 107.58	
Smoker/family member ratio			
0	7 (11.48)	68.40 ± 55.74	0.705
0.1 – 0.2	22 (36.07)	126.60 ± 145.75	
0.3 – 0.4	22 (36.07)	135.80 ± 149.16	
≥ 0.5	10 (16.39)	116.84 ± 94.46	
Father occupation			
Trader/seller in traditional market or stall	18 (29.51)	154.18 ± 168.44	0.747
Odd jobs	13 (21.31)	123.83 ± 156.06	
Laborer	8 (13.11)	82.64 ± 40.37	
Small entrepreneur	7 (11.48)	73.16 ± 41.27	
Drivers	5 (8.20)	132.48 ± 50.92	
Other occupation**	10 (16.39)	119.92 ± 125.09	
Mother occupation			
Homemakers	47 (77.05)	112.02 ± 131.51	0.369
Trader/seller in traditional market or stall	7 (11.48)	126.49 ± 117.71	
Other occupation***	7 (11.48)	192.92 ± 59.50	
Total	61 (100.00)	121.64 ± 131.48	

#Mean ± standard deviation

*Mann-Whitney test significant

**Foreman, security guard, blacksmith, retired

***Midwife, tailor, small entrepreneur

Table 2. Students' Urinary Cotinine Levels and The Presence of Smoker Family Members at Home

Urinary Cotinine Levels (ng/mL)	n (%)	Smoker Family Members at Home (n %)	
		Yes	No
< 10	7 (11.48)	6 (85.7)	1 (14.3)
10 – 100	39 (63.93)	34 (87.2)	5 (12.8)
100.1 – 200	5 (8.20)	4 (80.0)	1 (20.0)
> 200	10 (16.39)	10 (100)	0 (0)

Because the number of smoker family members and the total family members varied in each student's home, a ratio of smoker family members divided by the total family members was made to be able to state the relationship between the number of smoker family members and the intensity of SHS and THS exposure received by students, thus affecting urinary cotinine levels. The ratio obtained was of 0.0 to 0.8 (Table 2). However, Spearman's Rho test showed no correlation ($p=0.133$) between

the ratio and urinary cotinine levels. Students were divided into three groups based on their report card average marks to evaluate the effect of cigarette smoke exposure on learning performance (Table 3). This table shows the trend of students with report card average marks of ≤ 81.42 to have urinary cotinine levels ≥ 10 ng/mL. However, the chi-square test showed insignificant results ($p=0.390$). Similarly, the Spearman's Rho correlation showed a low and negative correlation ($r_s=-0.121$; $p=0.352$).

Table 3. Distribution of Urinary Cotinine Level and Students' Academic Performance

Urinary cotinine levels (ng/mL)	Number of Students Based on Academic Performance n (%)		
	71,08- 76,25	76,26- 81,42	81,43- 86,60
<10	4(57.1)	2(28.6)	1 (14.3)
10-100	11 (28.2)	24(61.5)	4 (10.3)
100,1-200	1 (20.0)	3 (60.0)	1 (20.0)
>200	6 (60.0)	3 (30.0)	1 (10.0)
Total	22 (36.1)	32 (52.4)	7 (11.5)

Because urinary cotinine levels were significantly higher in male students than female, the report card average marks were then separated by gender.

Figure 1 shows that the report card average marks in male students was lower than in female students, although it was not significant ($p=0.063$).

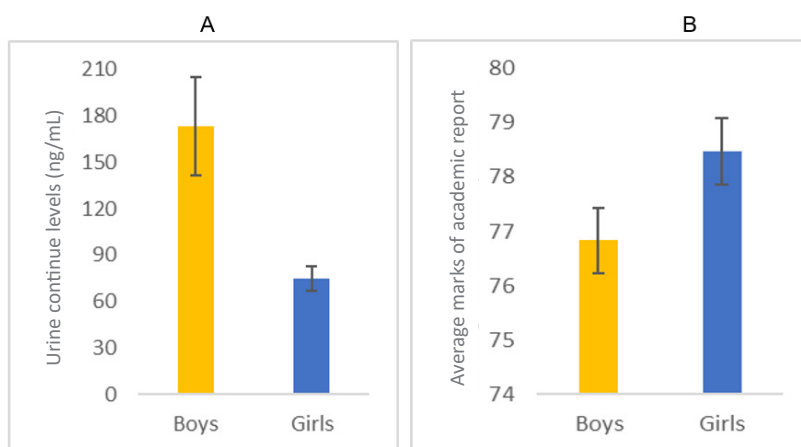


Figure 1. Urinary Cotinine Levels in Male Students were Significantly Higher than In Female Students (A). Boys Tended to Have Lower Academic Performance than Girls (B)

Discussion

Studies showed that 40% of children aged <15 years were exposed to SHS in their homes.^{10,11,16} SHS and THS exposures impact children's health, from upper and lower respiratory infections, asthma, otitis media, and cancers.^{1,10} SHS exposure can affect children's cognitive and behavioral performance. Nicotine could affect the brain regarding attention, memory, learning, and sleep disturbances.^{13,14} The ability to read and count was shown to decline in children aged 6-16 years who were exposed to SHS.¹⁷

This study showed that almost 90% of students stated that they had a family member who smoked at home. Smoking habits were also very likely to expose children with SHS and THS, such as smoking inside the house while meals and watching television with children and other family members, as evidenced by urinary cotinine levels on average of 121.64 ng/mL. Urinary cotinine levels can be used to differentiate between smokers, passive smokers, and non-smokers.¹⁸ However, due to differences in the technique for measuring urinary cotinine levels, to the best of the authors' knowledge, no fixed cut-off point is generally accepted to distinguish the three categories above. The consistent results showed that passive smokers (recipients of SHS and THS exposure from smokers) have significantly higher urinary cotinine levels than non-smokers. A study by Sharma et al.¹⁹ showed that passive smokers had cotinine levels on average 36.63 ± 64.57 ng/mL, significantly higher than non-smokers, namely 13.6 ± 12.73 ng/mL as measured using liquid chromatography and mass spectrometry. Susanto et al.²⁰ found that passive smokers had urinary cotinine levels (in median) of 30.1 ng/mL while non-smokers were 8.45 ng/mL, measured using the enzyme-linked immunosorbent assay (ELISA).

This study has not shown a significant relationship between students' urinary cotinine levels and the number of family members who smoke at home. This result might indicate that although the number of smoker family members at home was small, they intensively exposed children with SHS and THS due to bad smoking habits. Hence, the urinary cotinine showed a high level. Interestingly, urinary cotinine levels in boys were significantly higher than in girls. Boys have a tendency to play outside more and do not hesitate to join older age groups, such as teenagers and adult smokers, so that they may receive more SHS exposure than girls. Interview questions also missed asking about

the possibility of the students, especially boys, as active smokers because the study focused on the frame of children as passive smokers. Nevertheless, Riskesdas data revealed that the number of child smokers in Indonesia was still high, even increasing from 7.2% in 2013 to 9.1% in 2018.^{2,3}

The relationship between urinary cotinine levels and total student academic performance showed that the results were not significant. However, if the academic performance data were differentiated by gender, boys had lower academic performance than girls. Further confirmatory studies are needed to prove this because many factors determine academic performance. These results were in line with previous studies showing that exposure to SHS and THS can affect cognitive function.^{13,14,17}

Physiologically, healthy children aged 12-15 years followed by 7-11 years had better antioxidant capacity than younger age groups and adults.²¹ The high antioxidant capacity can neutralize oxidative metabolites from cigarettes that have the potential to damage cells, including neurons. This mechanism may explain why the impact of SHS and THS on cognitive function has not been obviously detected in this study. Nevertheless, consider that nicotine as the main component of cigarettes is not an oxidant and has direct effects on the brain, including the effects of addiction. Therefore, the main thing to take into account is the long-term impact of SHS and THS on cognitive function, especially the nicotine component, which may manifest in productive ages.¹⁴ The limitation of this study is that the assessment of academic performance through report cards alone to assess cognitive function seems inadequate. Thus, a more specific method is needed, such as an assessment of the level of intelligence, visuomotor integration, verbal memory, attention, and executive function with their respective standardized tests.²²

Conclusion

Children are very vulnerable to receiving SHS and THS exposure from smokers who unfortunately come from their own families. The smoking habits of family members are still very worrying. Hence it may promote long-term health impacts and reduce the quality of life of children at their productive age.

Acknowledgments

Authors would like to express a great appreciation to dr. Marhaen Hardjo, M.Biomed., PhD and dr. Gita Vita Soraya, PhD for their insights and comments that greatly improved this study.

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High Urinary Cotinine Levels and Low Academic Performance of Elementary School Students in Families with Careless Smoking Habits

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Submission date: 03-Feb-2023 08:38AM (UTC+0700)

Submission ID: 2005186955

File name: 2._AP1_199_197_203.pdf (264.85K)

Word count: 4156

Character count: 21653

Research Article

High Urinary Cotinine Levels and Low Academic Performance of Elementary School Students in Families with Careless Smoking HabitsIka Yustisia,^{1,2*} Nurdiana Sari,³ Suryani Tawali,⁴ Mutmainah Arif,⁵ Sitti Ramliah²¹Master Program of Biomedical Sciences, ²Department of Biochemistry,³Medical Study Program, ⁴Department Public Health and Preventive Medicine
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Received 6 April 2022; Accepted 8 December 2022

<https://doi.org/10.23886/ejki.10.199.197>**Abstract**

Children are the most vulnerable age group to become passive smokers due to cigarette smoke exposure, especially from family members. In addition, children who smoke passively are at high risk of experiencing declined cognitive performance. This study investigated the effect of cigarette smoke exposure on urinary cotinine levels and its relationship with elementary school students' academic performance. The observational study with a cross-sectional design was located in Pannampu Village, Tallo District, Kota Makassar. Interviews, collecting academic performance data, and measuring urinary cotinine levels using colorimetric techniques were carried out on elementary school students (n=77). The research activities took place from June to August 2018. This study found that 88.5% of students stated that they had family members who smoked at home with inappropriate habits of exposing children to cigarette smoke. Urinary cotinine levels showed an average of 121.64 ng/mL. All students with urinary cotinine levels above 200 ng/mL (16.4%) had smoker family members. The urinary cotinine levels in boys were higher (Mann-Whitney test, p=0.014) with lower report card average marks than in girls. The smoking habits of family members were still a concern and had not protected children from cigarette smoke exposure, as evidenced by the high average urinary cotinine levels, especially in boys, which might affect their academic performance.

Keywords: smoking habits, urinary cotinine, passive smoking.**Kadar Kotinin Urin dan Prestasi Belajar Siswa Sekolah Dasar pada Keluarga dengan Perilaku Merokok yang Buruk****Abstrak**

Anak-anak merupakan kelompok usia paling rentan menjadi perokok pasif akibat paparan asap rokok dari orang-orang sekeliling terutama anggota keluarga. Anak-anak perokok pasif berisiko tinggi mengalami gangguan kesehatan termasuk penurunan kemampuan kognitif. Penelitian ini bertujuan menganalisis pengaruh asap rokok terhadap kadar kotinin urin dan hubungannya dengan prestasi belajar anak. Penelitian observasional analitik dengan desain potong lintang ini berlokasi di Kelurahan Pannampu, Kecamatan Tallo, Kota Makassar. Wawancara, pengumpulan data prestasi akademik, dan pengukuran kadar kotinin urin dengan teknik kolorimetri dilakukan pada siswa usia sekolah dasar (n=77). Penelitian dilakukan pada bulan Juni-agustus 2018. Didapatkan 88,5% siswa memiliki anggota keluarga perokok di rumah dengan perilaku berisiko memajankan asap rokok pada anak. Kadar kotinin urin menunjukkan nilai rerata 121,64 ng/mL. Semua siswa dengan kadar konitin urin >200 ng/mL (16,4%) memiliki anggota keluarga perokok di rumah. Kadar kotinin urin pada anak laki-laki lebih tinggi (Mann-Whitney test, p=0,014) dengan rerata nilai rapor lebih rendah dibandingkan anak perempuan. Perilaku merokok anggota keluarga memprihatinkan dan belum melindungi anak dari pajanan asap rokok yang dibuktikan dengan rerata kadar kotinin urin yang tinggi terutama pada anak laki-laki. Diperlukan upaya yang kuat untuk melindungi anak dari bahaya rokok yang dapat bermanifestasi penurunan fungsi kognitif di masa depan.

Kata kunci: perilaku merokok, kotinin urin, perokok pasif.

Introduction

Cigarette smoking and other forms of tobacco consumption are public health problem that is part of behavioral health risk factors (HRFs).¹ Although many studies prove the health effects of cigarette smoking, education and campaigns are still being promoted; however, the proportion of smokers has not changed significantly. For example, data from Indonesian Basic Health Research (Riset Kesehatan Dasar/Riskesdas) showed that the proportion of smokers in Indonesia was 29.3% in 2013, only a slight decrease to 28.9% in 2018.^{2,3} In South Sulawesi Province, the proportion of smokers also decreased from 27.0% to 25.9%.⁴ However, these proportions are still high, considering that the global target for reducing smoking prevalence is 15.5% by 2025.¹

When a cigarette is burned, the smoke is formed, containing nicotine and other toxic chemical compounds that are harmful to health. Cigarette smoke inhaled by smokers is referred to as primary cigarette smoke (FHS) (first-hand smoke = FHS).⁵ Meanwhile, smoke from the burning end of a cigarette and is exhaled by smokers is called second-hand smoke (SHS).⁶ The smoke produced from burning cigarettes does not evaporate into the air but still contains nicotine residue that can pollute the air. It can react with other chemicals to form carcinogens and then stick to the surface of surrounding objects such as clothes, carpets, walls, furniture, bed linen, car dashboards, and children's toys. This latter type of cigarette smoke, referred to as third-hand smoke (THS), can remain in the environment for years.^{5,6} Nicotine and tobacco-specific nitrosamines were common contaminants found on many indoor surfaces, including floors, walls, and windows.^{7,8} Nicotine derived from THS can enter the body through the skin, respiratory tract, and digestive tract.⁹

Children are the most vulnerable group to receive SHS and THS from smokers around them. The main source of cigarette smoke exposure to children is from their homes and surroundings.^{10,11} Individuals exposed to SHS, including children are susceptible to receiving more than 250 carcinogenic compounds and hazardous chemicals.¹² SHS exposure to children is likely to cause premature death and increase the risk of developing acute respiratory infections, severe asthma symptoms, ear infections, and cancers.^{1,10} In addition, SHS exposure and most likely THS can affect brain development. Cause this organ is very sensitive to toxins.^{13,14} This study aims to evaluate the impact of SHS and THS exposure, especially the

nicotine component, on the academic performance of elementary school students. The intensity of nicotine exposure in children is determined by measuring the levels of the main metabolite of nicotine, namely cotinine, in urine.

Method

The research protocol has received ethical approval from the Ethics Commission of the Faculty of Medicine, Hasanuddin University (number 19/H.4.8.4.5.31/PP36-KOMETIK/ 2018). This analytic observational study with a cross-sectional design was conducted in Pannampu Village, Tallo District, Kota Makassar, centered on SD Inpres Pannampu III. The study began with education about the negative impact of smoking on children's health, then explained the research objectives and procedures, conducted informed consent, interviewed subjects, collected morning urine samples and academic performance data in the form of report cards. The activities took place from June to August 2018. The study population was elementary school students by taking a total sample of SD Inpres Pannampu III that met the inclusion criteria included 5th and 6th grade students, aged ten-year-old and over, received permission from their parents, proficient in reading and writing. Students were excluded if they decided to leave their willingness to be research subjects and were not cooperative during interviews and urine collection.

Interview

In the interview, subjects were asked to answer several questions including personal data (name, age, residential address, how many children are in the family, residential address, and class at school), family characteristics (number of family members, parents' occupations, number of family members outside the nuclear family, and number of families who smoke), and smoking habits and activities of family members.

Urine Collection and Cotinine Level Determination

Research subjects were asked to collect their morning urine into the pot after obtaining their parents' permission and signing the informed consent form. The urine samples were then picked up and transported to the Biochemistry Laboratory, Faculty of Medicine, Hasanuddin University using an icebox for cotinine levels measurement.

Cotinine levels in urine were measured using a colorimetric method modified from previous study.¹⁴

In summary, 0.5 mL of urine sample was added with 0.5 mL of ethanol, 0.2 mL of acetic buffer (4 M, pH 4.7), 0.1 mL of 1.5 M KCN, 0.1 mL of 0.44 M chloramine, and 0.5 mL of 78 mM barbituric acid. The solution mixture was incubated for 100 minutes at room temperature. The color formed from the solution was measured at a wavelength of 508 nm using a Genesys 150 UV VIS spectrophotometer (Thermo Scientific). Each sample was measured in triplicate. The absorbance value obtained was then converted to a concentration value based on the standard cotinine (Sigma C-016) curve, developed using the CurveExpert software.

Data Analysis

Data on academic performance used the average marks of report cards in the last two semesters. The data were collected and processed using Microsoft Excel 2016 software. Statistical analysis was carried out using the SPSS 20 for Windows.

Results

Out of 77 Grade 14 and Grade 6 students at SD Inpres Pannampu III who met the inclusion criteria, 61 students participated in the study until the end (Table 1). Sixteen students were uncooperative during the interview and did not collect urinary samples.

Smoking Habits of Family Members

The interview result showed several important points regarding the smoking habits of family members in the house (Figure 1). Of the 61 students interviewed, 54 students (88.5%) stated that they had at least one smoker family member at home and 7 students (11.5%) did not have smoker family members. From the students who had smoker family members, the smokers often smoked when

they were with the students (75.4%), when they gathered with the family (62.3%), while watching TV (68.9%) and after meals (52.4%). However, most students stated (73.8%) that other family members had advised smokers to stop or reduce cigarette consumption and not smoke inside the house.

Urinary Cotinine Level

The urinary cotinine levels (Table 1), both those who have family members who smoke at home and who did not have an average of 121.64 ng/mL ranging between 0.83 ng/mL to 655.78 ng/mL. This urinary cotinine level was not significantly different in the four age groups. Boys had significantly higher ($p=0.014$) level (173.20 ng/mL) than girls (74.90 ng/mL). Although not statistically significant, the greater the number of smoker family members and the higher the ratio of smokers per total number of family members at home, the higher the students' urinary cotinine level. Students whose fathers worked as traders in the traditional market or stalls, odd jobs, and laborers tended to have higher urinary cotinine levels. Students whose mothers work at home as homemakers have lower urinary cotinine levels than students whose mothers have certain professions such as traders/sellers in the traditional market or stalls, midwives, tailors, or small entrepreneurs.

The wide range of urinary cotinine levels (0.83 ng/mL – 655.78 ng/mL) was grouped into three as presented in Table 2. Most of the students ($n=39$; 63.93%) had urinary cotinine levels of 10 – 100 ng/mL and 87.2% of them had smoker family members. Students with cotinine concentrations \geq 200 ng/mL were a fairly high percentage (16.4%) and all of them had family members who smoked at home. So that there was a tendency for students who have smoker family members at home to have urinary cotinine levels higher than 10 ng/mL.

Table 1. Students' Urinary Cotinine Levels on Assessed Variables

Variables	n (%)	Urinary Cotinine Levels (ng/mL) [†]	p value
Ages (yo)			
10	21 (34.43)	132.32 ± 155.49	0.544
11	26 (42.62)	116.37 ± 142.54	
12	13 (21.31)	115.84 ± 59.22	
13	1 (1.64)	109.69	
Sex*			
Male	29 (47.54)	173.20 ± 171.81	0.014
Female	32 (52.46)	74.90 ± 45.35	
Number of family in the house			
4-6	28 (45.90)	141.81 ± 156.76	0.384
7-9	21 (34.43)	79.72 ± 47.64	
≥ 10	12 (19.67)	147.92 ± 157.18	
Number of smoker family member			
None	7 (11.48)	68.41 ± 55.74	0.688
1	27 (44.26)	130.28 ± 136.68	
2 - 4	20 (32.79)	133.69 ± 151.70	
≥ 5	7 (11.48)	107.09 ± 107.58	
Smoker/family member ratio			
0	7 (11.48)	68.40 ± 55.74	0.705
0.1 – 0.2	22 (36.07)	126.60 ± 145.75	
0.3 – 0.4	22 (36.07)	135.80 ± 149.16	
≥ 0.5	10 (16.39)	116.84 ± 94.46	
Father occupation			
Trader/seller in traditional market or stall	18 (29.51)	154.18 ± 168.44	0.747
Odd jobs	13 (21.31)	123.83 ± 156.06	
Laborer	8 (13.11)	82.64 ± 40.37	
Small entrepreneur	7 (11.48)	73.16 ± 41.27	
Drivers	5 (8.20)	132.48 ± 50.92	
Other occupation**	10 (16.39)	119.92 ± 125.09	
Mother occupation			
Homemakers	47 (77.05)	112.02 ± 131.51	0.369
Trader/seller in traditional market or stall	7 (11.48)	126.49 ± 117.71	
Other occupation***	7 (11.48)	192.92 ± 59.50	
Total	61 (100.00)	121.64 ± 131.48	

[†]Mean ± standard deviation

*Mann-Whitney test significant

**Foreman, security guard, blacksmith, retired

***Midwife, tailor, small entrepreneur

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 Table 2. Students' Urinary Cotinine Levels and The Presence of Smoker Family Members at Home

Urinary Cotinine Levels (ng/mL)	n (%)	Smoker Family Members at Home (n %)	
		Yes	No
< 10	7 (11.48)	6 (85.7)	1 (14.3)
10 – 100	39 (63.93)	34 (87.2)	5 (12.8)
100.1 – 200	5 (8.20)	4 (80.0)	1 (20.0)
> 200	10 (16.39)	10 (100)	0 (0)

Because the number of smoker family members and the total family members varied in each student's home, a ratio of smoker family members divided by the total family members was made to be able to state the relationship between the number of smoker family members and the intensity of SHS and THS exposure received by students, thus affecting urinary cotinine levels. The ratio obtained was of 0.0 to 0.8 (Table 2). However, Spearman's Rho test showed no correlation ($p=0.133$) between

the ratio and urinary cotinine levels. Students were divided into three groups based on their report card average marks to evaluate the effect of cigarette smoke exposure on learning performance (Table 3). This table shows the trend of students with report card average marks of ≤ 81.42 to have urinary cotinine levels ≥ 10 ng/mL. However, the chi-square test showed insignificant results ($p=0.390$). Similarly, the Spearman's Rho correlation showed a low and negative correlation ($rs=-0.121$; $p=0.352$).

Table 3. Distribution of Urinary Cotinine Level and Students' Academic Performance

Urinary cotinine levels (ng/mL)	Number of Students Based on Academic Performance n (%)		
	71,08- 76,25	76,26- 81,42	81,43- 86,60
<10	4(57.1)	2(28.6)	1 (14.3)
10-100	11 (28.2)	24(61.5)	4 (10.3)
100.1-200	1 (20.0)	3 (60.0)	1 (20.0)
>200	6 (60.0)	3 (30.0)	1 (10.0)
Total	22 (36.1)	32 (52.4)	7 (11.5)

Because urinary cotinine levels were significantly higher in male students than female, the report card average marks were then separated by gender.

Figure 1 shows that the report card average marks in male students was lower than in female students, although it was not significant ($p=0.063$).

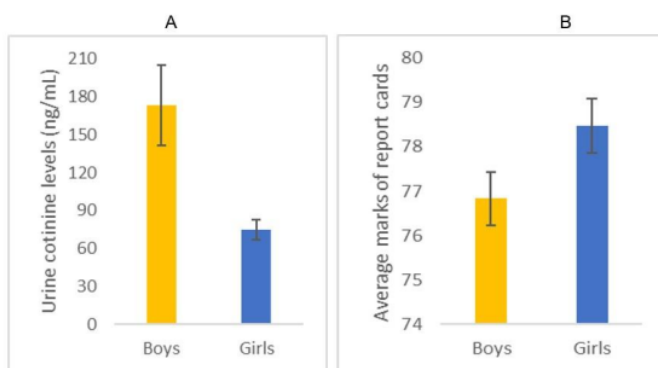


Figure 1. Urinary Cotinine Levels in Male Students were Significantly Higher than In Female Students (A). Boys Tended to Have Lower Academic Performance than Girls (B)

Discussion ⁹

Studies showed that 40% of children aged <15 years were exposed to SHS in their homes.^{10,11,16} SHS and THS exposures impact children's health, from upper and lower respiratory infections, asthma, otitis media, and cancers.^{1,10} SHS exposure can affect children's cognitive and behavioral performance. Nicotine could affect the brain regarding attention, memory, learning, and sleep disturbances.^{13,14} The ability to read and count was shown to decline in children aged 6-16 years who were exposed to SHS.¹⁷

This study showed that almost 90% of students stated that they had a family member who smoked at home. Smoking habits were also very likely to expose children with SHS and THS, such as smoking inside the house while meals and watching television with children and other family members, as evidenced by urinary cotinine levels on average 121.64 ng/mL. Urinary cotinine levels can be used to differentiate between smokers, passive smokers, and non-smokers.¹⁸ However, due to differences in technique for measuring urinary cotinine levels, to the best of the authors' knowledge, no fixed cut-off point is generally accepted to distinguish the three categories above. The consistent results showed that passive smokers (recipients of SHS and THS exposure from smokers) have significantly higher urinary cotinine levels than non-smokers. A study by Sharma et al.¹⁹ showed that passive smokers had cotinine levels on average 36.63±64.57 ng/mL, significantly higher than non-smokers, namely 13.6±12.73 ng/mL as measured using liquid chromatography and mass spectrometry. Susanto et al.²⁰ found that passive smokers had urinary cotinine levels (in median) of 30.1 ng/mL while non-smokers were 8.45 ng/mL, measured using the enzyme-linked immunosorbent assay (ELISA). This study has not shown a significant relationship between students' urinary cotinine levels and the number of family members who smoke at home. This result might indicate that although the number of smoker family members at home was small, they intensively exposed children with SHS and THS due to bad smoking habits. Hence, the urinary cotinine showed a high level. Interestingly, urinary cotinine levels in boys were significantly higher than in girls. Boys have a tendency to play outside more and do not hesitate to join older age groups, such as teenagers and adult smokers, so that they may receive more SHS exposure than girls. Interview questions also missed asking about

the possibility of the students, especially boys, as active smokers because the study focused on the frame of children as passive smokers. Nevertheless, Riskesdas data revealed that the number of child smokers in Indonesia was still high, even increasing from 7.2% in 2013 to 9.1% in 2018.^{2,3}

The relationship between urinary cotinine levels and total student academic performance showed that the results were not significant. However, if the academic performance data were differentiated by gender, boys had lower academic performance than girls. Further confirmatory studies are needed to prove this because many factors determine academic performance. These results were in line with previous studies showing that exposure to SHS and THS can affect cognitive function.^{13,14,17}

Physiologically, healthy children aged 12-15 years followed by 7-11 years had better antioxidant capacity than younger age groups and adults.²¹ The high antioxidant capacity can neutralize oxidative metabolites from cigarettes that have the potential to damage cells, including neurons. This mechanism may explain why the impact of SHS and THS on cognitive function has not been obviously detected in this study. Nevertheless, consider that nicotine as the main component of cigarettes is not an oxidant and has direct effects on the brain, including the effects of addiction.¹⁷ Therefore, the main thing to take into account is the long-term impact of SHS and THS on cognitive function, especially the nicotine component, which may manifest in productive ages.¹⁴ The limitation of this study is that the assessment of academic performance through report cards alone to assess cognitive function seems inadequate. Thus, a more specific method is needed, such as an assessment of the level of intelligence, visuomotor integration, verbal memory, attention, and executive function with their respective standardized tests.²²

Conclusion

Children are very vulnerable to receiving SHS and THS exposure from smokers who unfortunately come from their own families. The smoking habits of family members are still very worrying. Hence it may promote long-term health impacts and reduce the quality of life of children at their productive age.

Acknowledgments

Authors would like to express a great appreciation to dr. Marhaen Hardjo, M.Biomed., PhD and dr. Gita Vita Soraya, PhD for their insights and comments that greatly improved this study.

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