APPLICATION OF SOLID-PHASE MICROEXTRACTION FOR THE ANALYSIS OF POLYCYCLIC AROMATIC HYDROCARBONS IN HYOPHILA INVOLUTA AS A NATURAL BIOSENSOR

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ABSTRACT

This work describes a simple and rapid extraction method by using solid-phase microextraction (SPME) technique for the determination of polycyclic aromatic hydrocarbons (PAHs) from mosses of *Hyophila involuta* which is found commonly and abundantly throughout Peninsular Malaysia. Mosses have been chosen in our study since they are effective bioindicators as they take up nutrients and pollutants directly from the atmosphere and they have no root system. These properties make them ideal for evaluating pollutant levels in the air over a monitored period of time. This study has been developed and optimized based on the extraction of the analytes of interest from the headspace over the moss samples with SPME followed by the analysis by gas chromatography-flame ionization detector (GC-FID). Factors affecting the SPME method, such as extraction mode and effects of the addition of chemical modifiers were evaluated, and the precision, linearity and detection limits were validated. Extraction was performed using operating conditions with standard addition method using 100 µm poly(dimethylsiloxane) (PDMS) fiber under the following conditions: extraction temperature of 150 ºC, extraction time of 90 min, desorption time and temperature of 15 min and 275 ºC, respectively, with the addition of 250 µl of 5% (w/v) NaCl salt solution. The headspace SPME method was developed using non-spiked and spiked moss samples in a concentration range of 0-2600 ng/g. The method showed good linearity for the tested concentration range with regression coefficient ranging between 0.9721 and 0.9974. The relative standard deviations which were determined at the low (150 ng/g), mid (2000 ng/g) and high (2600 ng/g) concentrations for all PAHs were between 2-37%, except for some compounds (benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, and indeno(123-cd)pyrene) which exhibited slightly higher values (42-69%). Recoveries were in relatively high levels for all

compounds at three different concentration levels, and ranged from 80-119% except for a few compounds (benzo(k)fluoranthene, indeno(123-cd)pyrene, dibenzo(ah)anthracene, and benzo(ghi)perylene) which showed lower recoveries (23-67%) at concentration level of 150 ng/g. The detection and quantification limits were 1.3-193.1 pg/g and 4.2-643.6 pg/g, respectively for all PAH compounds studied. The developed method was then applied for the quantification of PAH compounds with heavy traffic pollution from contaminated moss samples collected from selected stations in the Klang Valley area.

ABSTRAK
(VERSI BAHASA MALAYSIA)

Kerja ini menerangkan satu kaedah pengekstrakan yang cepat and ringkas dengan mengunakan teknik solid-phase microextraction (SPME) untuk pentafsiran kehadiran polisiklik aromatik hidrokarbon (PAH) dalam lumut *Hyophila involuta* yang dijumpai secara umum dengan banyaknya di Semenanjung Malaysia. Lumut telah dipilih untuk dikaji kerana mereka adalah bioindikator yang berkesan dimana mereka mengambil nutrien dan polutan terus daripada atmosfera dan juga tidak mempunyai sistem akar. Sifat-sifat ini membolehkan lumut menilai peringkat polutan di dalam udara untuk suatu tempoh masa. Kajian ini telah dikembangkan dan dioptimumkan berdasarkan pengekstrakan analit-analit yang diminati daripada headspace atas sampel-sampel lumut dengan SPME diikuti oleh analisis kromatografi gas-pengesan pengionan nyala (GC-FID). Faktor-faktor yang mempengaruhi kaedah SPME, seperti mode pengekstrakan dan kesan daripada penambahan kimia pengubah telah dinilai, dan ketelitian, linieritas dan batas deteksi telah disahkan. Pengekstrakan telah dilakukan mengunakan keadaan operasi kaedah standard samping dengan 100 µm polydimethylsiloxane (PDMS) fiber dibawah keadaan-keadaan berikut: suhu pengesktrakan pada 150 ºC, masa pengekstrakan selama 90 min, masa dan suhu desorpsi selama 15 min dan 275 ºC masing-masing, dengan penambahan sebanyak 250 µl 5% (w/v) NaCl larutan garam. Kaedah headspace SPME telah dibangunkan dengan mengunakan sampel-sampel lumut yang tidak spike dan yang sudah spike dalam julat kepekatan 0-2600 ng/g. Kaedah ini menunjukkan linearitas yang baik untuk julat yang diuji dengan pekali regresi dalam lingkungan antara 0.9721 dan 0.9974. Relatif standard deviasi yang telah ditentukan pada kepekatan rendah (150 ng/g), sederhana (2000 ng/g) dan tinggi (2600 ng/g) untuk semua PAH adalah antara 2-37%, kecuali untuk beberapa sebatian (benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, and indeno(123-

cd)pyrene) yang mempamerkan nilai peratusan yang agak tinggi (42-69%). Recoveries adalah agak tinggi tahapnya untuk semua sebatian PAH bagi ketiga-tiga tahap kepekatan, dan berada dalam julat 80-119% kecuali untuk beberapa sebatian (benzo(k)fluoranthene, indeno(123-cd)pyrene, dibenzo(ah)anthracene, and benzo(ghi)perylene) yang mempamerkan recoveries yang rendah (23-67%). Batas pengesanan and kuantifikasi adalah dalam lingkungan 1.3-193.1 pg/g dan 4.2-643.6 pg/g masing-masing untuk semua sebatian yang diuji. Kaedah headspace SPME yang dikembangkan kemudian diguna untuk pengkuantifikasi sebatian-sebatian PAH dengan pencemaran lalu lintas yang padat daripada lumut-lumut yang nyata tercemar yang dikumpul daripada pilihan stesen-stesen di sekitar kawasan Lembah Klang.

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LIST OF ABBREVIATIONS

% w/v	percentage of weight/volume
∑PAHs	total PAHs content
Acpe	acenaphthene
Acpy	acenaphthylene
Ant	anthracene
AQI	Air Quality Index
BaA	benzo(a)anthracene
BaP	benzo(a)pyrene
BbFl	benzo(b)fluoranthene
BghiP	benzo(ghi)perylene
BkFl	benzo(k)fluoranthene
Chr	chrysene
CO	carbon monoxide
CO_2	carbon dioxide
DBA	dibenzo(ah)anthracene
DI	direct immersion
DSASE	direct sonication-assisted solvent extraction
Fl	fluoranthene
Flu	fluorene
GC	gas chromatograph
GC-FID	gas chromatograph-flame ionization detector
GC-MS	gas chromatograph-mass spectrometry
HPLC	high performance liquid chromatography
HS	headspace

InP	indeno(123-cd)pyrene
K_{fs}	distribution constant
KL	Kuala Lumpur
LDR	linear dynamic range
LOD	limit of detection
LOQ	limit of quantitation
MAE	microwave extraction
MTBE	methyl *tert*-butyl ether
NaCl	sodium chloride
Nap	naphthalene
NO_2	nitrogen dioxide
OCP	organochlorine pesticides
P_c	critical pressure
PA	polyacrylate
PAH	polycyclic aromatic hydrocarbon
PCB	polychlorinated biphenyl
PDMS	poly(dimethylsiloxane)
PDMS-DVB	poly(dimethylsiloxane-divinylbenzene)
Phe	phenanthrene
PJ	Petaling Jaya
PLE	pressurized liquid extraction
PM	particulate matter
$PM_{2.5}$	measure of particles with a diameter of less than 2.5 or equal to a nominal 2.5 micrometers
PM_{10}	measure of particles with a diameter of less than 10 or equal to a nominal 10 micrometers

POP	persistent organic pollutant
PSI	Pollution Standards Index
Pyr	pyrene
R^2	correlation coefficient
RSD	relative standard deviation
SO_2	sulphur dioxide
SOC	semi-volatile organic compound
SFE	supercritical fluid extraction
SPE	solid-phase extraction
SPME	solid-phase micro-extraction
SWE	subcritical water extraction
T_c	critical temperature
USE	ultrasonic extraction
U.S. EPA	United States Environmental Protection Agency
VOC	volatile organic compound