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Assessing Smoking Behaviour and Tobacco Smoke Exposure: Definitions and Methods*

by

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SUMMARY

In recent years, the increased availability of tobacco products other than conventional cigarettes, the use of puffing topography devices for smoking behaviour studies and the use of biomarkers to study smoke constituents exposure have generated the need for a more comprehensive set of definitions concerning smoking behaviour and exposure to smoke. The definitions offered in this paper are based on many years of practical experience and on consensus within a broad group of scientists working in these areas. It is intended that, with wider and more consistent usage, these definitions should reduce any misunderstandings and facilitate interpretation of future studies. [Beitr. Tabakforsch. Int. 25 (2013) 685–699]

ZUSAMMENFASSUNG

In den letzten Jahren stieg die Verfügbarkeit von gegenüber konventionellen Zigaretten neuen und veränderten Tabakprodukten sowie die Verwendung von Geräten zur Erfassung der Topographie des Rauchmusters in Raucherverhaltensstudien und der Einsatz von Biomarkern zur Messung der Exposition gegenüber Rauchbestandteilen. Diese Entwicklung hat eine umfassendere Zusammenstellung von Definitionen bezüglich des Rauchverhaltens und der Rauchexposition notwendig gemacht. Die in diesem Artikel beschriebenen Definitionen basieren auf jahrelanger Erfahrung und dem Konsens einer Gruppe von Wissenschaftlern, die auf diesem Gebiet arbeitet. Es ist beabsichtigt und erwünscht, dass die breite und konsistente Anwendung dieser Definitionen zu weniger Missverständnissen und einer verbesserten Verständlichkeit zukünftiger Studien führt. [Beitr. Tabakforsch. Int. 25 (2009) 685–699]

RESUME

Ces dernières années, une plus grande disponibilité des produits du tabac autres que les cigarettes conventionnelles, l'utilisation d'appareils permettant de mesurer la topographie des bouffées lors d'études sur le comportement du fumeur et l'utilisation de biomarqueurs pour mesurer l'exposition aux composés de la fumée ont suscité le besoin d'avoir un ensemble plus complet de définitions concernant le tabagisme et exposition à la fumée. Les définitions proposées dans ce document sont basées sur de nombreuses années d'expérience et sur le consensus au sein d'un large groupe de scientifiques travaillant dans ces domaines. Il est prévu que, avec une utilisation plus large et plus cohérente, ces définitions devraient réduire les malentendus et faciliter l'interprétation des études futures. [Beitr. Tabakforsch. Int. 25 (2013) 685–699]

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INTRODUCTION

The process of smoking a cigarette is highly variable. For any given smoker, each puff on a single cigarette may vary and each cigarette may be smoked differently from occasion to occasion. For groups of smokers, the same product may be smoked differently, even if measurements are taken at a single time point. The frequency of smoking and the intensity of puffing and inhalation are parameters that can be measured at both the individual and group level. Together with many other parameters, such individual and group smoking characteristics are called "smoking behaviour".

Smoking behaviour has been studied for over 100 years but, even in very early studies, a major focus was on how aspects of smoking behaviour alter the exposure to smoke constituents, such as nicotine (1, 2), and to smoke particulates (3). Studies now cover all parameters that can be measured as components of smoking behaviour (4, 5). In recent years, smoking behaviour studies have included new products other than conventional cigarettes, such as cigarettes that heat rather than burn tobacco (6–8) or electronic cigarettes (e-cigs) (9) and, in this paper, the term "smoking article" is used to include them.

The wide variation in individual and group smoking behaviour causes difficulties in determining the exact amount of exposure to any smoke constituent but such information is essential for the scientific assessment of the use of smoking articles. Many approaches have been used to quantify human smoke exposure, including estimates based on:

- The number of cigarettes smoked per day (CPD) or over a longer period of time using the Brinkman index (10, 11) and pack years (PY) (12, 13);
- Smoking topography (14–18), including puffing pattern (19–21), inhalation pattern (22), mouth and pulmonary retention rates (4, 23), to improve the accuracy of intake and uptake measurements (24);
- Smoke duplicators, to mimic human smoking patterns on smoking machines (24);
- Used cigarette filter analysis, for human smoking yields (24, 26, 27); and
- Biomarkers of exposure in body fluids, for uptake (28–33).

The INTERNATIONAL ORGANIZATION FOR STANDARDIZATION (ISO) has produced a standard vocabulary for tobacco terms (34) but this does not cover many aspects of human use of the products. Therefore, BAKER *et al.* (35) proposed a set of definitions for cigarette smoking behaviour to be applied to smoke exposure studies.

After these definitions were advanced, papers on other aspects of smoking behaviour and smoke exposure were published. These other papers addressed possible regulatory aspects of tobacco products based on smoking behaviour and exposure studies as outlined by the WORLD HEALTH ORGANIZATION STUDY GROUP ON TOBACCO PRODUCT REGULATION (TobReg) reports (36, 37) and those of the INSTITUTE OF MEDICINE of the USA (IOM) (31, 38). Further, the availability of hand-held smoking recording devices (5, 39–41) has facilitated the gathering of data on the puffing aspects of smoking behaviour and smoke exposure. It is clear from these sources that the original terms described by BAKER *et al.* (35) are no longer comprehensive and require the addition of new terms to address current knowledge of smoking behaviour and smoke exposure measurement.

The aim of this paper is to define the terms and methods used for assessing smoking behaviour and exposure, with the objective of generating a more uniform application of the terms used by scientists working in this field of research.

SMOKE EXPOSURE TERMS

An alphabetical list of all the terms included in this paper along with common synonyms is shown in Table 1, with the preferred term shown in plain text and the synonym in italics. Table 1 should be used as an index for the terms that are listed in Table 2. In Table 2, the definitions and comments are separated loosely into three categories: 'Smoking behaviour', 'Smoking topography' and 'Biomarkers'. The definitions are split into these categories mainly to group the related terms for ease of location, even though it is recognised that the split is arbitrary as smoking topography is a subset of smoking behaviour. For each definition, units of measurement are given, along with an example and, typically, a comment on the method or its application.

In Table 2, most examples are given in terms of cigarettes because they are the most widely used tobacco products and the great majority of scientific publications on smoking behaviour and constituent exposure report on use of cigarettes. The terms that are concerned with puffing behaviour apply to both machine and human smoking situations. The difference between puffing and inhalation is highlighted intentionally as they are distinct processes.

Some of the terms defined in Table 2 have more than one application; for example, 'mean flow' can be used to describe the values for a single puff obtained from a puffing topography instrument or it can be used to describe the arithmetic mean value obtained across all puffs from a smoking article. Similarly, 'peak flow' may be the peak flow rate per puff or the peak flow rate across all puffs from a single smoking article. The definitions allow for either circumstance and authors are urged to specify carefully the precise application in their work.

The term 'puff number' is used herein to refer to the total count of puffs taken from a smoking article rather than specifying a particular puff in a topographic record. This is consistent with the ISO standard (34). Annotated graphics showing how some of these terms relate to a typical puffing topography profile and a typical smoking topography profile are given in Figures 1 and 2.

DISCUSSION

Puffing and inhalation

The intake of smoke includes distinct physical activities. In a first step, the smoker puffs on the smoking article and the smoke is drawn into the mouth. The soft palate at the back of the mouth remains closed, sealing the oral cavity (42). For some smokers, the smoke is expelled directly from the

Table 1. Alphabetical list of terms and synonyms in smoke exposure and smoking behaviour studies. In this table, the preferred term is given in plain text and alternative terms in *italic text*.

Term	Definition in Table 2
Amount absorbed [See 'uptake']	15
Average flow [See 'mean flow']	18
Biologically effective dose (BED)	42
Biomarker of exposure (BOE)	41
Brinkman index (BI)	1
Cigarettes per day (CPD)	2
Daily consumption [See 'cigarettes per day']	2
Dose [See 'exposure']	3
Dropped puffs	16
Exhalation	4
Exhalation duration	5
Exhalation volume	6
Exposure	3
Human smoking yield (HSY)	7
Individual puff draft pressure [See 'puff pressure']	32
Individual puff draw pressure [See 'puff pressure']	32
Inhalation	8
Inhalation depth [See 'inhalation volume']	10
Inhalation duration	9
Inhalation volume	10
Intake (I)	11
Inter-puff interval [See 'puff interval']	30
Internal dose [See 'uptake']	15
Machine-derived yield [See 'machine yield']	12
Machine yield (MY)	12
Maximum dose [See 'exposure']	3
Mean draft pressure [See 'mean draw pressure']	17
Mean draw pressure	17
Mean flow	18
Mean puff duration	19
Mean puff interval	20
Mean puff volume	21
Mean resistance	22
Mouth hold	23
Mouth level exposure (MLE) [See 'human smoking yield']	7
Mouth spill (MSp)	24
Pack years (PY)	13
Partial puffs [See 'sub-puffs']	38
Peak draft pressure [See 'peak draw pressure']	25
Peak draft resistance [See 'peak resistance']	27
Peak draw pressure	25
Peak flow	26
Peak resistance	27
Puff count [See 'puff number']	31
Puff duration	28
Puff frequency	29
Puff interval	30
Puff number	31
Puff pressure	32
Puff to puff interval [See 'puff interval']	30
Puff volume	33
Puffing pattern [See 'puffing topography']	34
Puffing topography	34
Pulmonary retention [See 'retention']	14
Retention (R)	14
Smoking duration	35
Smoking machine yield [See 'machine yield']	12
Smoking topography	36
Smoulder time	37
Sub-puffs	38
Time between puffs [See 'puff interval']	30
Total puff duration	39
Total puff interval [See 'smoulder time']	37
Total puff volume	40
Uptake (U)	15
Yield [See 'machine yield']	12
Yield in-use (YIU) [See 'human smoking yield']	7

mouth and no inhalation step occurs. This is a typical smoking topography pattern observed in many cigar smokers. For many cigarette smokers, the smoker then relaxes the soft palate in less than a second after the end of the puff. This may lead to a second step, smoke inhalation, in which the smoker draws the smoke into the lungs by a conscious, voluntary movement. The second step requires additional air to be drawn into the respiratory tract either through the mouth or the nasal passage and this air mixes with the smoke during the process of inhalation (4, 5). Thus, puffing volume and inhalation volume are separate parameters that are not linked. Any given smoker may take a large puff with no, or minimal, subsequent inhalation. The converse is also possible; that is, a small puff volume followed by a large inhalation volume may be observed. This point is often overlooked in research reports measuring puffing without the measurement of subsequent inhalation.

Portable puff recording instruments

A number of recent research reports have used portable smoking instruments to record puffing topography measurements. Typically, they measure parameters of puffing behaviour such as puff number, puff interval, peak flow and mean flow. However, they are not capable of measuring other parameters such as mouth hold, mouth spill and any parameters associated with inhalation. Therefore, these instruments only give estimates of human smoking yield, but not an entire smoking behaviour topography profile. As noted above, there is no direct relationship between puffing parameters and subsequent inhalation, and therefore, measurements of inhalation and exhalation are also required to complete the smoking topography profile (41, 43). Nonetheless, subject to these limitations, these portable puffing topography instruments are extremely useful in field studies (i.e., away from a labo-

Table 2. Definition of terms, units and comments.

No.	Term	Definition	Units and methods	Example and comment (see footnote for abbreviations)
			Smoking behaviour	
1	Brinkman index (BI)	Daily number of cigarettes × years smoked	No units (index) Recall or daily records / logs	• 600 [20 CPD × 30 years] Used as a crude surrogate for lifetime exposure in epidemiological studies (10, 11)
2	Cigarettes per day (CPD)	Number of cigarettes smoked per day	CPD Can be assessed by interview or questionnaire Other methods are diaries, collection of used cigarette filters	• 20 CPD Used as a crude surrogate for mean daily exposure without allowing for constituent machine yield and smoking behaviour (71)
3	Exposure	Concentration × time for a specified constituent present in the external medium (such as air, water, tobacco smoke, food)	Mass per mass of external medium × time Mass per volume of external medium × time Mass per unit × units per time (mg, µg, ng, pg) × (min, h, d, wks, yrs) Also assessed by measuring biomarkers of exposure in body fluids or organs (biological monitoring)	• Daily exposure of smokers to nicotine from cigarettes = Human Smoking Yield $_{[nic]}$ × CPD Exposure may be thought of as the maximum potential dose of the smoke constituent. Intake and uptake cannot exceed exposure
4	Exhalation (post-puffing)	The act of expelling inhaled and puffed material from the respiratory space and mouth or nasal passages of a smoker	See 'Exhalation duration' and 'Exhalation volume'	
5	Exhalation duration	Time from end of inhalation until the end of the exhalation phase of the breathing cycle	Time (s) from start of expiratory flow until the end of this action	• 5 s Typically measured by flow monitoring through the oral cavity with the nasal passage pinched Other methods include respiratory inductive plethysmography devices such as bands or vests, which record chest movements (4) Note: if no inhalation has occurred, the duration of the expulsion of puffed material from the oral cavity can be measured
6	Exhalation volume	Volume of material expelled from the respiratory space after a puff and inhalation from a smoking article	Volume (mL) expired from the start of expiratory flow until the end of this action	• 500 mL Typically measured by flow monitoring through the oral cavity with the nasal passage pinched Other methods include respiratory inductive plethysmography devices such as bands or vests, which record chest movements (4) Note: if no inhalation has occurred, the volume of puffed material expelled from the oral cavity can be measured

Table 2. contd.

No.	Term	Definition	Units and methods	Example and comment (see footnote for abbreviations)
			Smoking behaviour (contd.)	
7	Human smoking yield (HSY)	Amount of a given mainstream smoke constituent exiting the smoking article into the mouth, when a given person smokes that product	Mass (mg, µg, ng, pg) Measured by duplication of the human smoking profile in a smoking machine; or by used cigarette filter analysis techniques	• mg NFDPM / cig • mg nicotine / cig • µg acrolein / cig HSY varies across individuals and usually is different from MY (27, 72)
8	Inhalation (post-puffing)	The act of moving puffed material from the mouth into the trachea and respiratory space of a smoker	See 'Inhalation duration' and 'Inhalation volume'	This is a separate action from puffing (43)
9	Inhalation duration	Time from start of inhalation until start of exhalation phase of the breathing cycle	Time (s) Measured by respiratory inductive plethysmography using bands or vests, which record chest movements	• 5 s This time does not include the initial puff or any mouth-hold period (43)
10	Inhalation volume	Volume of inspiration into trachea and respiratory space	Volume (mL) Measured by respiratory inductive plethysmography using bands or vests, which record chest movements The volume may be expressed as a percent of the vital capacity for each subject	• 500 mL • 11% vital capacity See references (5, 43)
11	Intake (I)	The mass or fraction of puffed material or of a specific smoke constituent that is taken into the mouth or into the respiratory system, prior to any deposition or absorption	Mass (mg, µg, ng, pg)	• mg NFDPM / cig • mg NFDPM / puff • mg nicotine / cig • µg acrolein / cig Intake is similar to human smoke yield
12	Machine yield (MY)	Amount of a given constituent exiting the smoking article under machine smoking conditions at a specified smoking regime, e.g., International Organization for Standardization (ISO), Health Canada, Massachusetts	Mass (mg, µg, ng, pg) Trap on glass fibre filter (Cambridge filter) or suitable solid or liquid traps Quantitate with common analytical techniques (e.g., gravimetric, photometric, GC, HPLC)	• mg NFDPM / cig • mg nicotine / cig • µg acrolein / cig MY is a standard measurement and usually is different from HSY (73)
13	Pack years (PY)	Number of years smoking × number of packs of cigarettes smoked per day	A composite number (packs per day × years of smoking) Subject recall or diary records/ logs	• 20 PY = One pack per day for 20 years or half a pack per day for 40 years or 2 packs per day for 10 years (based on a pack of 20 cigarettes) Used as a crude surrogate for lifetime exposure without allowing for constituent MY and smoking behaviour, often used in epidemiological studies (12) although this has been criticized (13)

Table 2. contd.

No.	Term	Definition	Units and methods	Example and comment (see footnote for abbreviations)
			Smoking behaviour (contd.)	
14	Retention (R)	Difference between the amount of a smoke constituent inhaled (intake) and the amount exhaled over subsequent breathing cycles	Mass (% retained) Volumes are measured as described for inhalation and exhalation parameters. Mass is estimated by appropriate physical or chemical analysis	• µg solanesol / cig • mg nicotine / cig • n % nicotine retained / puff For many smoke constituents, e.g., nicotine, R is practically identical with the amount absorbed but for others, e.g.,some PAH, R does not equal absorption (4)
15	Uptake (U)	The amount of smoke constituent which is absorbed through the mucosa of the mouth, respiratory tract and lung	Mass (mg, µg, ng, pg) Cannot be directly measured in a human but is deduced indirectly from the level of the compound (or metabolite) measured in body fluids or organs	• mg nicotine / cigarette • mg nicotine / puff • mg nicotine / day After absorption, a local tissue accumulation or a systemic distribution may occur
			Smoking topography	
16	Dropped puffs	Puffs not counted toward the puff number, if their volume is below a specified amount	Number of puffs below a pre-defined volume Measured on a puffing topography instrument	• 4 puffs less than 5 mL
17	Mean draw pressure	The average mouth-end pressure (vacuum) per puff for each puff or across all puffs from a smoking article	mm H_2O Measured with a puffing topography instrument	• 225 mm H_2O
18	Mean flow	The average volumetric flow rate for each puff or across all puffs from a smoking article	mL / s Measured with a puffing topography instrument	• 32.4 mL / s
19	Mean puff duration	The average time from start to end of a puff, across all puffs from a smoking article	Time (s)	• 2.4 s / puff
20	Mean puff interval	The average time between the end of one puff and the start of the next puff, across all puffs from a smoking article	Time (s)	• 41 s
21	Mean puff volume	The average volume across all puffs from a smoking article	Volume (mL)	• 42.5 mL / puff

Table 2. Contd.

No.	Term	Definition	Units and methods	Example and comment (see footnote for abbreviations)
			Smoking topography (contd.)	
22	Mean resistance	The average of the ratio of mean draw to mean flow across all puffs from a smoking article	mm H_2O / (mL / s)	• 6.9 mm H_2O / (mL / s) Often used to indicate the amount of effort that a smoker must make to draw a puff from a smoking article
23	Mouth hold	The time over which a puff is held in the oral cavity before inhalation or exhalation	Time (s) Measured by observation, typically with a smoking topography device or by difference between the end of a puff and the start of inhalation measured as described above	• 1 s
24	Mouth spill (MSp)	Amount of smoke or smoke constituent that is spilt from the mouth after puffing and not inhaled by the smoker	Mass per puff Mass per smoking article (mg, µg, ng, pg) No established method	MSp is observed in many smoking behaviour studies and it affects exposure, intake and uptake but no standard method for quantification has been devised (4)
25	Peak draw pressure	The maximum mouth end pressure (vacuum) for each puff or across all puffs from a smoking article	mm H_2O Measured with a puffing topography instrument	• 360 mm H_2O
26	Peak flow	The maximum volumetric flow rate for each puff or across all puffs from a smoking article	mL / s Measured with a puffing topography instrument	• 44.3 mL / s The highest peak flow for any puff from a smoking article is usually described as the maximum flow per article
27	Peak resistance	The highest ratio of peak draw to peak flow across all puffs from a smoking article	mm H_2O / (mL / s) Calculated from measurements made with a puffing topography instrument	• 7.1 mm H_2O / (mL / s)
28	Puff duration	Time from start to end of a specified puff	Time (s)	• 1.65 s / puff Puff duration is fixed in standard machine smoking regimes (e.g., 2 s for the ISO method) (73)
29	Puff frequency	Number of puffs taken in a fixed amount of time	Number per time interval	• 2 / min Puff frequency is fixed in standard machine smoking regimes (e.g., 1 / min for the ISO method) (73)
30	Puff interval	Time between the end of a specified puff and the start of the next puff	Time (s)	• 43 s Puff interval is fixed in standard machine smoking regimes (e.g., 58 s for the ISO method) (73)

Table 2. Contd.

No.	Term	Definition	Units and methods	Example and comment (see footnote for abbreviations)
			Smoking topography (contd.)	
31	Puff number	The count of the total number of puffs per smoking article	Number per smoking article Taken from the puffing topography record	• 7 puffs / cigarette As defined in ISO 10185 (33)
32	Puff pressure	The pressure (vacuum) recorded per individual puff from a smoking article	mm H_2O Measured with a puffing topography instrument	• 244 mm H_2O
33	Puff volume	Amount of smoke and air drawn into the mouth during an individual puff	Volume (mL)	• 42.1 mL / puff Puff volume is fixed in standard machine smoking regimes (e.g., 35 mL for the ISO method) (73)
34	Puffing topography	The profile of puff characteristics including puff frequency, duration, volume, interval, and regularity of these parameters for a smoking article	A complex set of measurements including puff frequency (per minute) with a comment on puff interval Other units are described in related definitions Puffing topography is assessed by specialised instruments with pressure and flow measurement capabilities	• 2 / min with regular puff interval [See Figure 1] Puff pattern is fixed, with regular puff intervals in standard machine smoking regimes (e.g., 1 / min with a 58 s puff interval for the ISO method) (73)
35	Smoking duration	Total elapsed time from the start of the first puff to the end of the last puff of a smoking article	Time (s)	• 390 s
36	Smoking topography	The complete pattern using a smoking article, including puffing, mouth hold, inhalation and exhalation	A complex set of measurement parameters based on definitions included herein Smoking topography is assessed by two or more specialised instruments. The puffing topography [see above] is supplemented with separate measurements of inhalation and exhalation	[See Figure 2]
37	Smoulder time	Smoking duration minus the sum of individual puff durations for a single smoking article. This also equals the sum of puff intervals for that smoking article	Time (s)	• 373 s

Table 2. contd.

No.	Term	Definition	Units and methods	Example and comment (see footnote for abbreviations)
			Smoking topography (contd.)	
38	Sub-puffs	The count of contiguous puffs within a pre-specified time interval, prior to the combination of data for analysis as a single puff	Number Recorded on a puffing topography instrument	• 3 sub-puffs A concatenated sub-puff is one of a group of puffs for which the start of the following puff occurs within a specified time interval from the end of the previous puff (e.g., 0.8 s) [See Figure 1] There are no sub-puffs in standard machine smoking regimes
39	Total puff duration	Cumulative time for all puffs from a smoking article	Time (s)	• 16.5 s / cigarette
40	Total puff volume	Combined volumes for all puffs from a smoking article	Volume (mL)	• 425 mL / cigarette
			Biomarkers	
41	Biomarker of exposure (BOE)	A smoke constituent or its metabolite that is measured as a concentration in body fluids, excreta or tissues (e.g., blood, urine, saliva, exhaled air, hair, sweat). BOE may also be measured as protein or DNA adducts	Mass per unit volume (body fluid) Mass per mg creatinine (urine sample) Mass per 24h (total urinary collection) Mass per mass other marker (e.g.,tissues, urine, exhaled breath) Measured with a validated method	• 15.5 mg nicotine equivalents / 24h (urine) (48) • 220 ng cotinine / mL (plasma) (55) • 25 ppm CO (exhaled air) (74) • 362 ng NNAL / 24h (urine) (48) • 260 pg NNAL / mg creatinine (urine) (48) • 1.8 mg HPMA / 24h (urine) (54) • 500 ng HPMA / mg creatinine (urine) (75) • 3.3 µg MHBMA / 24h (urine) (54) • 59.7 ng MHBMA / mg creatinine (urine) • 4.1 µg SPMA / 24h (urine) (54) • 18.4 ng / mg creatinine (urine) (75)
42	Biologically effective dose (BED)	Amount of a smoke constituent or metabolite that is bound to a macromolecule (e.g., protein, DNA, RNA) of a specific tissue or organ	Mass per mass (tissues) Mass per mass (protein- or DNA- adducts) Measured with a validated method	• 92 fmol HPB / mg DNA (lung of smokers) (76) • 0.042 fmol BaP adducts / mg albumin (plasma or serum) (77) • 9.9 adducts per 10^9 nucleotides (urothelial cell DNA, often expressed as a RAL) (78) • 4.6 % COHb (74)

Footnote: Abbreviations used and not defined within the table:

BaP: Benzo[*a*]pyrene
CO: Carbon monoxide
COHb: Carboxyhaemoglobin
DNA: Deoxyribose nucleic acid
GC: Gas chromatography
HPB: 4-Hydroxy-1-(3-pyridyl)-1-butanone [a compound released from NNK or NNN adducts upon hydrolysis]
HPLC: High performance liquid chromatography
HPMA: 3-Hydroxypropyl-mercapturic acid [a metabolite of acrolein]
MHBMA: Monohydroxybutenyl-mercapturic acid [a metabolite of 1,3-butadiene]
NFDPM: Nicotine-free dry particulate material
NNAL: 4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanol
NNK: 4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone
NNN: *N'*-Nitrosonornicotine
RAL: Relative adduct level [the ratio of the number of adducted nucleotides to the number of nucleotides with adducts]
RNA: Ribose nucleic acid
SPMA: *S*-Phenylmercapturic acid [a metabolite of benzene].

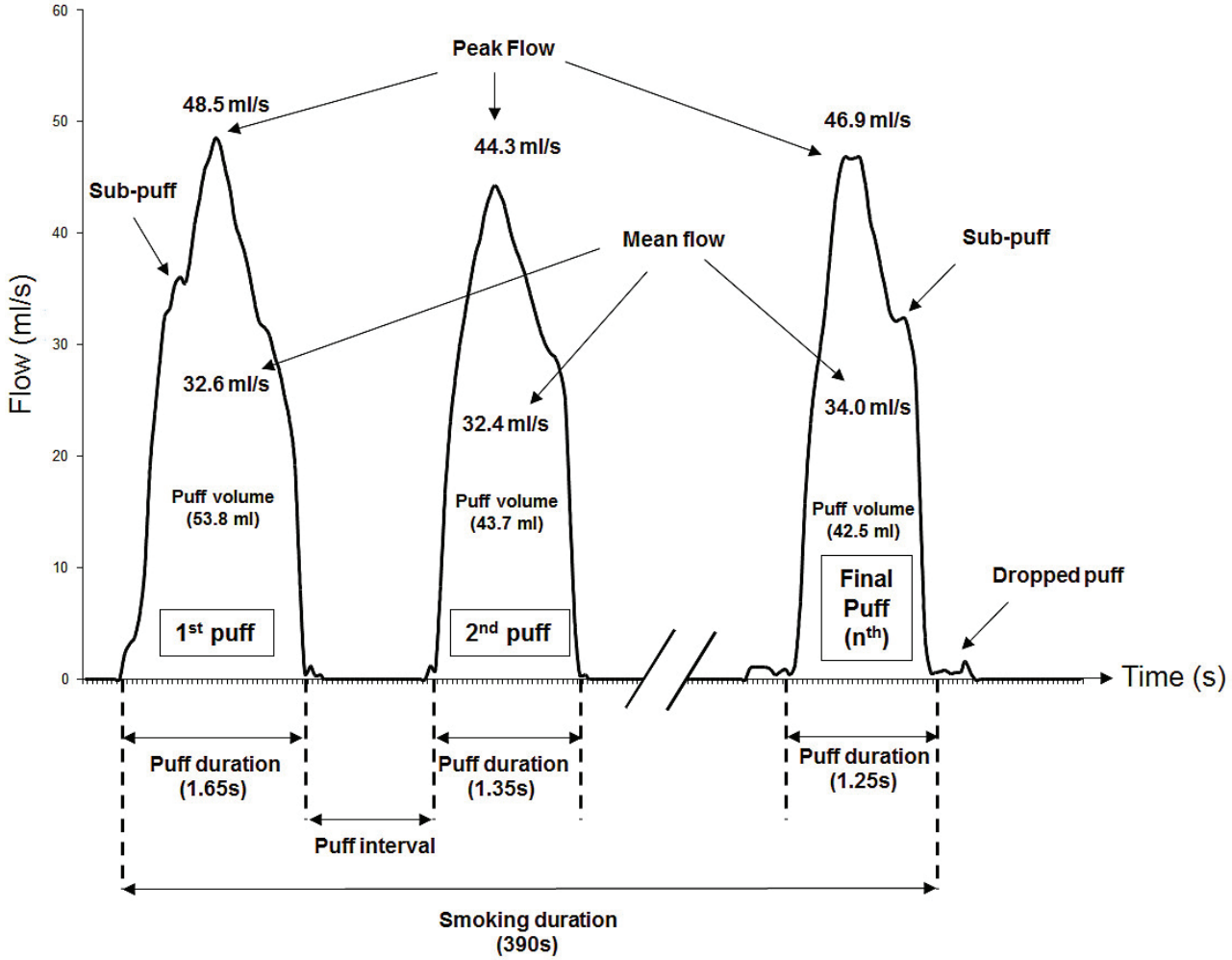

Figure 1. An example of a puffing topography trace.

ratory environment) and they can be used easily in cross-sectional studies of puffing behaviour. They measure fewer parameters of human smoking but produce data more rapidly than those obtained with more-labour intensive, laboratory-based, puffing duplication techniques (25). Further, smoking behaviour is affected by the setting of the investigation (25) and the use of a portable instrument may allow a smoker to interact with a smoking article in a more typical manner than a laboratory environment.

Intake, uptake and retention

These terms and their definitions cover subtly different aspects of exposure to smoke. For intake, our definition refers to the amount of a smoke constituent that is taken into the mouth. This usage is consistent with the approach of others, in separate disciplines such as risk assessment (44, 45) and chemical safety assessment (46), where intake is defined as a maximum exposure, prior to an absorption step. Even if smoke is expelled from the mouth immediately after puffing, a portion of the amount of any smoke constituent's intake may be retained in the oral cavity. In a similar manner, if an inhalation step occurs, then, after exhalation, a portion of any smoke constituent may be retained in the respiratory tract. This constituent retention may occur by a number of processes; for example, particle deposition onto a mucosal surface or by absorption into the tissue or bloodstream (5). Uptake is used to define the absorption of smoke constituents, which is distinct from retention. For some smoke constituents, such as nicotine, retention and uptake are practically identical (47) but for others, particularly hydrophobic constituents in the smoke particulate phase, retention and uptake can be quite different. The topic of retention has been considered in more detail in a review by BAKER and DIXON (4).

Biomarkers

The use of biomarkers to estimate exposure to smoke constituents offers the potential to estimate smoke constituent uptake, without having to measure smoking behaviour. A number of reports, including those from the WORLD HEALTH ORGANIZATION TobReg group (35, 36) and those of the IOM, which were sponsored by the FOOD AND DRUG ADMINISTRATION (FDA) of the USA (30, 31, 38), urged the development of this area. Several recent studies have reported biomarker measurements (48–54). However, it was noted that the definitions used for biomarkers were not applied uniformly across these studies (38). The use of a biomarker for exposure assessment is not straightforward, for several reasons. First, the biomarker chosen should be the smoke constituent of interest itself or

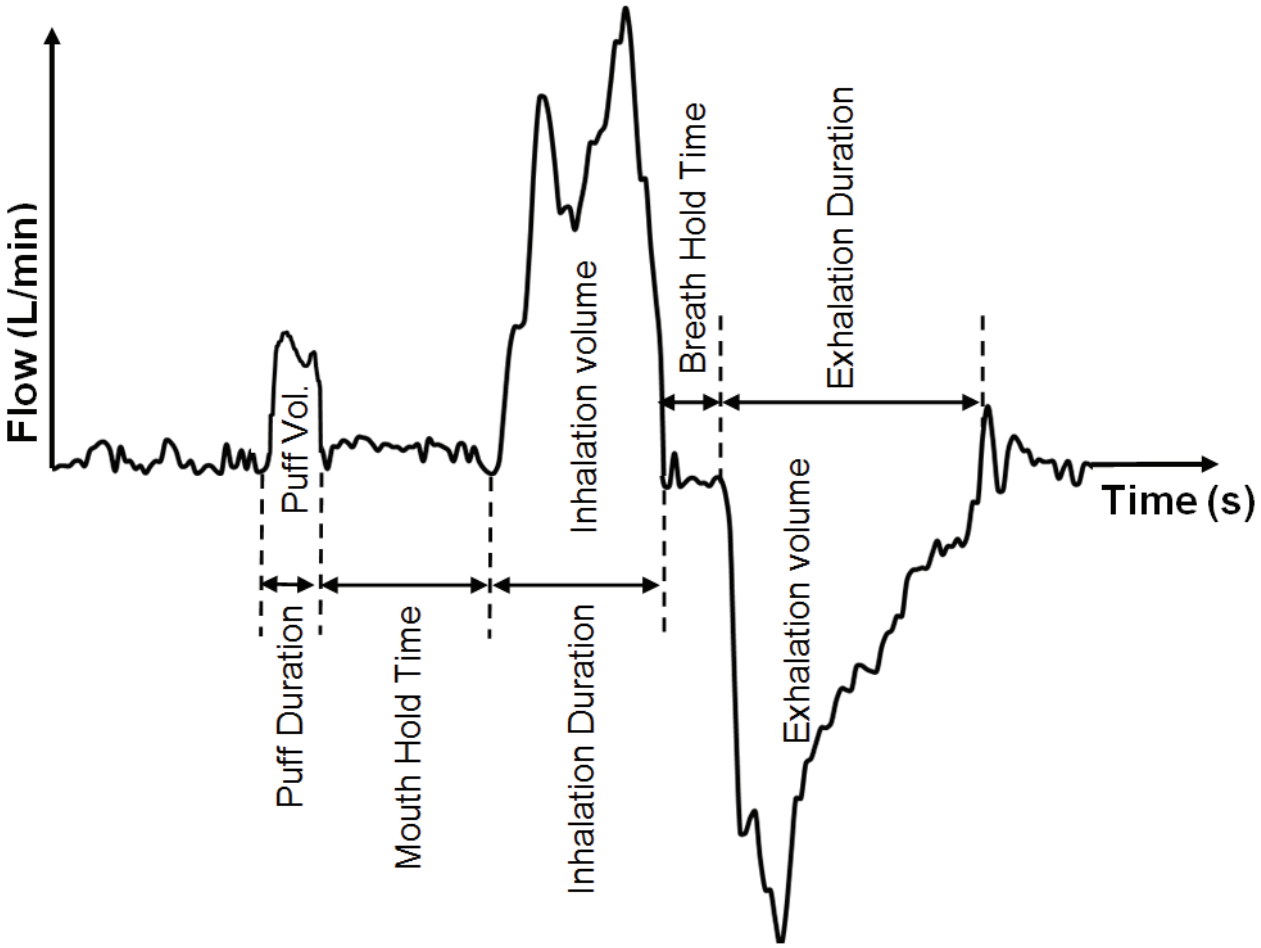

Figure 2. An example of a smoking topography trace. This profile was generated with a BIBO (Breathe In, Breathe Out) instrument (79). A BIBO instrument works on similar principles to the puffing topography instruments, but differs in that it has bi-directional pressure transducers which measure inhalation flow through the mouth and subsequent exhalation flow (with the nose clamped).
A BIBO instrument can be used to collect information on the volume of smoke taken into the mouth, the volume drawn into the airways and then exhaled. This enables researchers to measure the topography of the complete smoking cycle.

a well-characterised metabolite. When such a biomarker is available, the degree of polymorphism within the metabolic pathway should also be understood because fast- and slow-metabolisers of the same smoke constituent could give different biomarker measurements in body fluids following a similar exposure. This is why, even in the case of relatively abundant smoke constituents such as nicotine, the parent molecule plus five metabolites (48) or the parent plus up to nine metabolites (55, 56) are typically measured to estimate uptake. Second, metabolism may vary depending on the route of exposure to the tobacco constituent; for example, inhalation and respiratory absorption compared to buccal and gastrointestinal absorption following oral exposure (55). Third, the assays used for biomarker measurement must be fully validated and, although validation criteria are available (57–59), standard reference materials typically are not. Thus, results obtained from different testing laboratories cannot be directly compared, unless formalised inter-laboratory comparisons have been made (60). Fourth, the availability of a biomarker and an assay does not automatically qualify it for all uses (58, 59, 61). For example, a biomarker that shows a clear difference between smokers and non-smokers might be suitable to confirm abstinence from tobacco use, depending on the elimination half-life of the biomarker and the duration of the intervention study, but this does not mean that it is suitable for comparison between smokers of different products. As a practical example, measurement of nicotine plus metabolites can confirm smoking abstinence in the previous 2–3 days but it would not confirm any change in exposure to other constituents if, over recent days, a smoker switched from a conventional cigarette to a (theoretical) product generating lower overall human smoke constituent yields, but for which the nicotine biomarker levels were equivalent (62). In such a case, only chemical-specific biomarkers for the smoke constituents of interest would be suitable to confirm reduced uptake. For example, the exposure to benzo[*a*]pyrene in a modified tobacco product cannot be inferred from the measurement of nicotine and its metabolites, unless a relationship between these constituents has been established for that product (63). Furthermore, unless the relationships between uptake, metabolism,

distribution and excretion pathways are known across all polycyclic aromatic hydrocarbons of interest, they cannot be inferred from surrogate measurements.

In this paper, we have deliberately limited our description of biomarkers to those concerned with exposure. Numerous other categories have been proposed in the literature, including biomarkers of effect (64), biomarkers of potential harm (48), biomarkers of risk (65, 66), biomarkers of susceptibility (67–70) and surrogate biomarkers for these processes (38), often with overlapping descriptions. In the absence of clarity about their use, it would be premature to offer definitions for these categories of biomarkers. Nonetheless, the caveats about using validated assays for their measurement and the need for qualification before use in cross-sectional comparative studies, as noted above for the biomarkers of exposure, also apply to other biomarker categories.

Even the exposure biomarker category of 'biomarkers of effective dose', for which a definition is proposed, remains open to misinterpretation. The term 'biologically effective dose' is widely used based on the consideration that many toxicants (in particular carcinogens) require metabolic activation to the reactive chemical, which then interacts with DNA to form DNA adducts or with proteins to form protein adducts. The biologically effective dose, therefore, is a measure of the extent of exposure to and metabolic activation of a smoke constituent, but does not necessarily imply that any biological effect will result from it.

CONCLUSIONS

From the studies highlighted in this short paper, it is apparent that several recent developments have generated the need for a more comprehensive set of definitions concerning smoking behaviour and exposure to smoke. The definitions offered in this paper are based on many years of practical experience and on consensus within a broad group of scientists working in these areas. It is intended that, with wider and more consistent usage, these definitions should reduce any misunderstandings and facilitate interpretation of future studies.

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