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The Significance of Breath Sampling Frequency
on the Mouth Alcohol Effect

by
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ARTICLES

THE SIGNIFICANCE OF BREATH SAMPLING FREQUENCY ON THE MOUTH ALCOHOL EFFECT¹

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ABSTRACT

Retention of mouth alcohol may result in falsely high breath alcohol concentrations and is thus a concern in evidential breath alcohol testing. Frequent breath sampling used in the typical mouth alcohol experiment may underestimate the duration and magnitude of the mouth alcohol effect in comparison to the real-life forensic situation. On each of three separate trials, nineteen female, and eleven male, alcohol-free subjects rinsed their mouths with 20 mL of diluted vodka (20% alcohol v/v) for 20 seconds and then expectorated. On each of the trials, the subjects provided breath samples into an Intoxilyzer[®] 5000C either every two minutes, every four minutes, or every eight minutes, respectively. The subjects did not talk or open their mouths throughout the experiment except for providing the breath sample. The final breath result displayed, as well as whether or not the instrument detected mouth alcohol, was recorded. As expected, an increase in the frequency of breath sampling significantly decreased the magnitude and duration of the mouth alcohol effect. At eight minutes after rinsing the mouth with alcohol, the mean mouth alcohol concentration (MAC) (\pm SEM) was 0.072 (\pm 0.011) g/210 L, 0.057 (\pm 0.005) g/210 L and 0.041 (\pm 0.004) g/210 L for breath sampling every eight, four, and two minutes, respectively. At sixteen minutes after rinsing the mouth with the diluted vodka, a positive MAC was found in 47%, 33%, and 13% of the subjects for breath sampling every eight, four, and two minutes, respectively. The rate of detection of mouth alcohol by the Intoxilyzer[®] 5000C increased from 53% at MACs < 0.050 g/210 L to 100% at MACs > 0.199 g/210 L. Frequent breath sampling leads to a more rapid and greater decrease in the mouth alcohol effect. The ability of correctly detecting mouth alcohol with the Intoxilyzer[®] 5000C increases with increasing MAC.

RÉSUMÉ

L'alcool résiduel provenant de la cavité buccale peut mener à des résultats faussement élevés lorsque les concentrations d'alcool sont obtenus à l'aide d'échantillons d'haleine pour des fins judiciaires. La fréquence d'échantillonnage de spécimens d'haleine durant des protocoles expérimentaux typiques visant à étudier ce phénomène peut sous-estimer la durée et l'ampleur de l'effet résiduel de l'alcool de la bouche en comparaison avec des situations réelles. Pour chacune des trois expériences, dix neuf femmes et onze hommes qui s'étaient abstenus de tout alcool au préalable, se sont rincés la bouche avec 20mL de vodka diluée à 20% v/v pour 20 secondes et l'ont expectoré. Des échantillons d'haleine ont par la suite été obtenus des sujets et analysés à l'aide de l'Intoxilyzer[®] 5000C à des intervalles de 2 minutes, 4 minutes ou 8 minutes selon le protocole expérimental. On n'a pas permis aux

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sujets de parler ou d'ouvrir la bouche durant les différentes expériences sauf pour la collection des échantillons d'haleine. Le résultat final affiché sur l'instrument ainsi que l'observation d'alcool buccal décelé par l'instrument ont été inscrits. Tel qu'anticipé, une augmentation de la fréquence de collection d'échantillons d'haleine a diminué de façon significative l'ampleur et la durée de l'effet résiduel d'alcool de la bouche. Huit minutes après que les sujets se sont rincés la bouche avec de l'alcool, les concentrations moyennes d'alcool ("MAC" \pm écart type) étaient de 0.072 ± 0.011 g/210L, 0.057 ± 0.005 g/210L et 0.041 ± 0.004 g/210L lorsque l'échantillonnage a été fait à toutes les 8, 4 ou 2 minutes respectivement. Seize minutes après le rinçage de la bouche avec du vodka dilué, une "MAC" positive a été observée pour 47%, 33% et 13% des sujets selon que l'échantillonnage avait été obtenu à toutes les 8, 4 ou 2 minutes respectivement. L'affichage d'alcool buccal par l'Intoxilyzer® 5000C a augmenté de 53% lorsque les "MACs" étaient moindres de 0.050 g/210L à 100% lorsque les "MACs" étaient supérieures à 0.199 g/210L. La fréquence de collection d'échantillons d'haleine mène à une diminution plus grande et plus rapide de l'effet résiduel de l'alcool buccal. La capacité de déceler correctement de l'alcool résiduel de la bouche à l'aide de l'Intoxilyzer® 5000C augmente avec l'augmentation des concentrations "MAC".

INTRODUCTION

Retention of alcohol in the oral cavity from recent consumption of an alcoholic beverage, or alcohol-containing hygiene products (e.g. mouthwashes, breath sprays), or alcohol-containing medications, or regurgitation of stomach contents that contain a high alcohol concentration, may lead to an elevated breath alcohol concentration (1-4). Such an event has the potential to interfere with an evidential or a screening breath alcohol test. In order to prevent this from occurring, various precautionary measures have been implemented in evidential breath alcohol testing such as a fifteen-minute deprivation time and duplicate breath sampling (5). In addition, evidential breath alcohol testing instruments such as the Intoxilyzer®5000C have mouth alcohol detection systems (6).

As the mouth alcohol effect is a commonly raised defence in criminal courts, numerous studies have been conducted in this area to quantify this phenomenon (1-4, 7-22). However, most mouth alcohol experiments have not been conducted under forensic conditions. A typical mouth alcohol experiment involves an alcohol-free subject who rinses or gargles an alcohol solution in the mouth, expectorates, and then provides frequent breath samples into an instrument. This is clearly different from the usual drinking and driving situation in which a drinking subject consumes an alcoholic beverage, rather than gargles with it, and then provides a breath sample sometime later. These experiments, however, have been the basis of the 15- to 20- minute deprivation periods, which are a safeguard against mouth alcohol affecting the breath alcohol test. The relevance of such testing protocol should be examined.

The effect of rinsing versus swallowing has been determined in an earlier study (18), but the effect of frequency of breath testing has not been studied. The only reference to breath sampling frequency (i.e. the more frequent the exhalation the greater the volume of breath exhaled) is the Caddy *et al.* study (1). This study found that the greater expiration volume required for the "IR Intoxilyzer" resulted in a greater reduction in the mouth alcohol effect as compared to the "GC AlcoAnalyzer". However, the authors neither indicate the expiration volume required for each instrument nor attempt to quantitate this effect.

If a higher frequency of breath sampling leads to a faster dissipation of alcohol from the oral cavity, then the numerous experiments conducted under conditions of frequent breath sampling may underestimate the duration and magnitude of the mouth alcohol effect in real life forensic situations.

The main goal of the present study was to determine the significance of breath sampling frequency on the mouth alcohol effect. Additionally, the effectiveness of the Intoxilyzer[®] 5000C mouth alcohol detection system was also examined.

METHOD

On each of three trials, nineteen female and eleven male subjects who were alcohol-free, rinsed their mouths for 20 seconds with 20 millilitres (mL) of vodka diluted to 20% v/v alcohol, and then expectorated. The subjects did not have dentures or any other oral appliances in their mouths. The subjects then provided breath samples (a complete exhalation) into an Intoxilyzer[®] 5000C (CMI Inc., Owensboro, KY, USA) an Approved Instrument in Canada, for breath alcohol testing, that determines the alcohol concentration using infrared light (6). Breath samples were collected at one frequency for each trial. The sampling interval for each of the three trials was once every two minutes, once every four minutes, or once every eight minutes, respectively. To minimize potential confounding factors, such as evaporation of alcohol from the oral cavity, the subjects kept their mouths closed between breath samples. The spit trap mouthpiece was changed after each breath sample. The final result displayed on the digital display of the Intoxilyzer[®] 5000C was recorded, as well as whether or not the instrument detected mouth alcohol. The instrument detects mouth alcohol using a slope detector and indicates detection by displaying the message INVALID SAMPLE (19).

The Intoxilyzer[®] results, or MACs, are reported in this study as grams of alcohol in 210 litres of breath (g/210 L). This unit, which is widely used in the United States, is equivalent (when multiplied by 1000) to a blood alcohol concentration (BAC) measured in milligrams of alcohol in 100 millilitres of blood, when using the forensically acceptable blood to breath ratio of 2100 to 1. The **Criminal Code of Canada** and the *Recommended Standards and Procedures of the Canadian Society of Forensic Science Alcohol Test Committee* use these units (mg/100 mL) to define BACs, whether they are obtained from blood or breath samples (5). The accuracy of the Intoxilyzer[®] 5000C was checked before and after each sequence of testing on each subject using a breath alcohol simulator (Model 34C, Guth Laboratories Inc., Harrisburg, PA, USA) and all results were within $\pm 5\%$ of the target value of 0.100 g/210 L. The Intoxilyzer[®] records all breath alcohol concentrations < 0.007 g/210 L as zero.

The effect of sampling frequency on the magnitude of the mouth alcohol effect was analysed using a one-way ANOVA (Analysis of Variance) with sampling intervals (two, four, and eight minutes) as a within subject factor. The Tukey-Kramer Multiple Comparisons Test was used to determine the differences between treatment means at eight and 16 minutes.

RESULTS AND DISCUSSION

Figures 1 to 3 show the mean MAC [\pm Standard Error of the Mean (SEM)] obtained from the 30 alcohol-free subjects, at two-, four-, and eight-minute sampling intervals, respectively after the diluted vodka was expectorated. Figure 1 (breath sampling every two minutes) shows the typical exponential MAC curve obtained with frequent breath sampling. With fewer data points (Figure 2 and especially Figure 3) the smooth exponential curve becomes less apparent. All three experimental conditions include two common sampling times, at eight and 16 minutes after expectoration, and for that reason these times were used for a detailed examination of the effect of sampling frequency on the mouth alcohol effect.

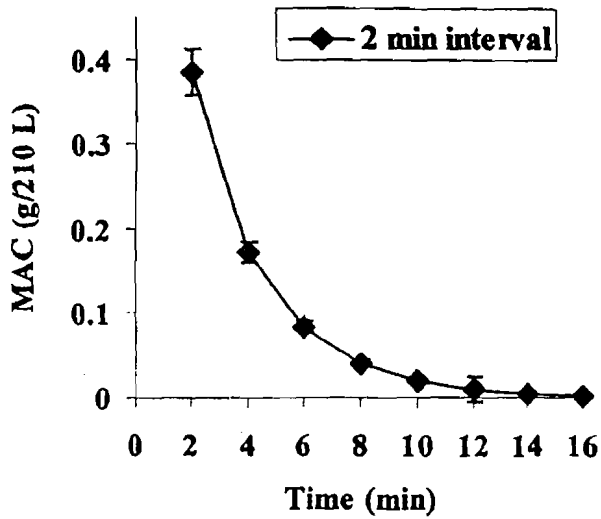


Figure 1. The mean MAC (\pm SEM) for 30 alcohol-free subjects after expectoration of the alcohol solution for breath sampling intervals of two minutes.

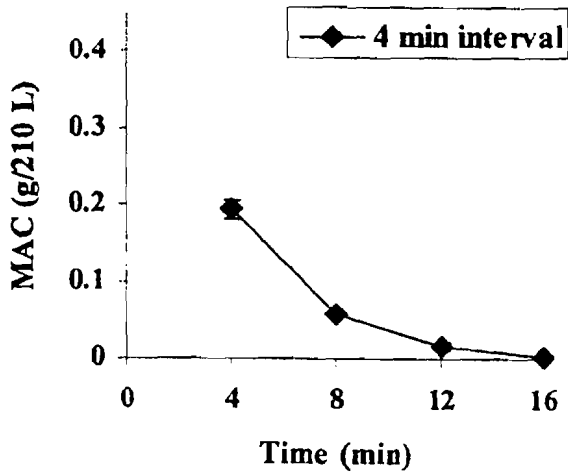


Figure 2. The mean MAC (\pm SEM) for 30 alcohol-free subjects after expectoration of the alcohol solution for breath sampling intervals of four minutes.

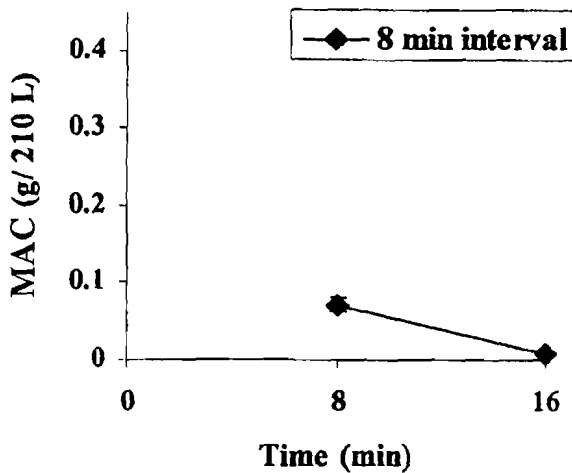


Figure 3. The mean MAC (\pm SEM) for 30 alcohol-free subjects after expectoration of the alcohol solution for breath sampling intervals of eight minutes.

TABLE 1

The mean MAC (\pm SEM) and the range (g/210 L) eight minutes after rinsing the mouth with diluted vodka in 30 alcohol-free subjects for breath sampling frequencies of every two minutes, every four minutes, and every eight minutes. The mean percent reduction from the least frequent breath sampling (every eight minutes) is also calculated.

Breath Sampling Frequency (Number of breath samples provided before and including at eight minutes)	Mean MAC (\pm SEM)	Range of MACs	Mean Percent (%) Decrease in Mean MAC
Every 2 minutes (4)	0.041 (0.004)	0.007–0.09	43
Every 4 minutes (2)	0.057 (0.005)	0.015–0.132	21
Every 8 minutes (1)	0.072 (0.011)	0.018–0.325	–

Table 1 shows the mean MACs (\pm SEM) at eight minutes after rinsing the mouth with the alcohol solution for breath sampling frequencies of every two minutes (or four exhalations), every four minutes (two exhalations) or every eight minutes (one exhalation), the range of MACs obtained, and the mean percentage decrease. As expected, the more frequent the sampling interval (which results in more exhalations which would remove alcohol from the oral cavity), the lower the MAC. The mean MACs decreased 21% (from 0.072 to 0.057 g/210 L) when the breath sampling frequency was doubled from once every eight minutes to once every four minutes. The mean MAC decreased 43% (from 0.072 g/210 L to 0.041 g/210 L) when the breath sampling frequency was increased four-fold (i.e. from once every eight minutes to once every two minutes). One-way ANOVA showed a significant effect of sampling frequency on MAC [$F(2,58) = 10.7$, $p=0.001$] obtained at eight minutes after expectoration. Post hoc analysis for differences between the treatment means revealed that the mean MAC obtained with the two-minute sampling interval was significantly less than the mean MAC obtained with the eight-minute sampling interval. However there were no significant differences between the mean MACs obtained with the two- and four-minute sampling frequencies, and the four- and eight-minute sampling frequencies.

Table 2 shows the mean MACs (\pm SEM) at 16 minutes after rinsing the mouth with alcohol. Here again the same pattern was observed, i.e. as the sampling frequency increased the mouth alcohol effect decreased. Sixteen minutes after the alcohol was rinsed in the mouth, the mean MAC decreased from 0.007 g/210 L (breath frequency every eight minutes) to 0.004 g/210 L (breath frequency every four minutes) to 0.001 g/210 L (breath sampling frequency every two minutes). Similar to the results obtained at eight minutes after expectorating, a one-way ANOVA showed a significant effect of sampling interval on MAC [$F(2,58) = 10.1$, $p=0.0002$]. Post hoc analysis for differences between the treatment means showed that the mean MAC using the two-minute sampling interval was significantly less than the mean MAC obtained with the eight-minute sampling interval. Again there were no significant differences in the mean MACs obtained with the two- and four-minute, and the four- and eight-minute sampling interval treatments.

As the mean MACs are low and there are a large number of zero MAC results at sixteen minutes, the percent reduction in MAC was not calculated. Instead, Table 2 shows the percentage of subjects who had a positive MAC at 16 minutes (or > 0.007 g/210 L as determined by the Intoxilyzer® 5000C). Forty-seven percent of the subjects had a positive MAC for breath sampling frequency every eight minutes, which decreased to 33% for

TABLE 2

The mean MAC (\pm SEM) and the range (g/210 L) 16 minutes after rinsing the mouth with diluted vodka in 30 alcohol-free subjects for breath sampling frequencies of every eight minutes, every four minutes, and every two minutes. The number of subjects with a positive BrAC and percent is also calculated.

Breath Sampling Frequency (Number of breath samples provided before and including at 16 minutes)	Mean BrAC (\pm SEM)	Range of MACs	Number of subjects with a positive MAC and %
Every 2 minutes (8)	0.001 (0.0007)	0.000–0.012	4 (13%)
Every 4 minutes (4)	0.004 (0.001)	0.000–0.019	10 (33%)
Every 8 minutes (2)	0.007 (0.002)	0.000–0.045	14 (47%)

breath sampling frequency every four minutes and 13% for breath sampling frequency every two minutes.

The typical mouth alcohol experiments cited earlier employ conditions that limit their applicability to actual “forensic” situations. Firstly, this study shows that frequent breath sampling causes the mouth alcohol effect to be of shorter duration and lesser magnitude (an average decrease of 21% to 43% at eight minutes after expectorating). Therefore, data collected under such experimental conditions (i.e., frequent sampling) may underestimate the mouth alcohol effect when applied to actual breath alcohol testing in the field.

Secondly, Wigmore and Leslie (18) found that rinsing causes the mouth alcohol effect to be of longer duration and magnitude than when an alcohol solution is swallowed. They showed that compared to swallowing, five minutes after an alcohol solution was rinsed in the mouth, the mean MAC was reduced from 0.091 g/210 L to 0.036 g/210 L (60% reduction). This reduction can be explained by a decrease in the area of the oral cavity exposed to the alcohol solution when swallowing compared to rinsing (19, 20).

As indicated earlier, the main purpose of this study was to determine the effect of breath sampling frequency on the typical mouth alcohol experiment, which is why the subjects rinsed their mouths with the alcohol solution. It is not intended to simulate an actual “forensic” situation. In a “forensic” situation, the subjects would have to swallow the alcohol solution and would be allowed to talk. A recent study on the effect of swallowing beer on alcohol-free subjects found that the BrAC caused by the mouth alcohol effect was only 0.011 g/210 L (range 0.000 – 0.013 g/210 L) after five minutes and all subjects had a zero MAC after ten minutes (21). In addition, Gullberg (14) has found that the effect of mouth alcohol on a drinking subject with a pre-existing breath alcohol concentration is of shorter duration and magnitude. More research needs to be conducted in this area.

Another aspect of this study was to determine the effectiveness of the Intoxilyzer[®] 5000C to detect mouth alcohol. Table 3 shows the number of times that mouth alcohol was detected compared to the MAC, and the percentage detection rate. Figure 4 shows that data as a bar graph. Generally, as the MAC increased, the percent of times that the Intoxilyzer[®] 5000C detected it increased. At MACs between 0.007 and 0.049 g/210 L the Intoxilyzer[®] detected mouth alcohol 53% of the time, which increased to 90% at MACs between 0.100 and 0.149 g/210 L. At MACs of 0.200 g/210 L and above, the Intoxilyzer[®] detected mouth alcohol 100% of the time. To our knowledge, this is the first published study which demonstrates that the effectiveness of the Intoxilyzer[®] 5000C mouth alcohol detector

TABLE 3

The range of MACs (g/210 L), the total number of results in that MAC range, the number that the Intoxilyzer® 5000C detected as mouth alcohol, and the number not detected as mouth alcohol. In addition the percent detection rate was calculated.

MAC Range (g/210 L)	Total Number	Number Detected as Mouth Alcohol	Number Not Detected as Mouth Alcohol	Percent Detection Rate
0.007-0.049	159	84	75	53
0.050-0.099	69	64	5	93
0.100-0.149	21	19	2	90
0.150-0.199	25	24	1	96
0.200+	47	47	0	100
Totals	321	238	83	74

Detection of Mouth Alcohol

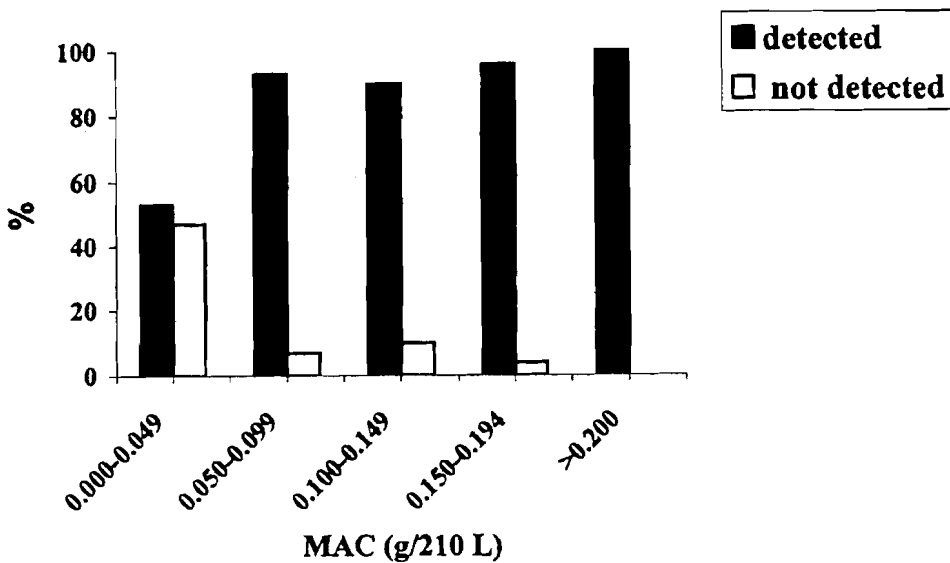


Figure 4. The percentage of times that the Intoxilyzer did not detect (open bars) or did detect (shaded bars) mouth alcohol with increasing MAC.

varies with MAC. Nevertheless, the combination of a relatively effective (at higher MACs) instrumental mouth alcohol detector with pre-existing safeguards, i.e., 1) 15 minutes alcohol-free period prior to the first test, 2) 15 minutes alcohol-free period between tests, and 3) the congruence of the breath test results, is an effective mechanism for avoiding interference by mouth alcohol.

CONCLUSIONS

The results of the present study show that as the breath sampling frequency increases, the duration and magnitude of the mouth alcohol effect decreases. This demonstrates that unrealistic experimental designs may limit the forensic applicability of conclusions in many mouth alcohol studies. This study also shows that the effectiveness of the mouth alcohol detector of the Intoxilyzer® 5000C increased with increasing MAC. The Intoxilyzer® 5000C is 100% effective in detecting mouth alcohol at MACs of 0.200.

g/210 L and higher. It is suggested that forensic alcohol experts carefully evaluate the experimental conditions used in the mouth alcohol studies when applying the results to actual forensic situations.

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