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Evaluation of methyl *tert*-butyl ether (MTBE) as an interference on commercial breath-alcohol analyzers☆

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Abstract

Anecdotal reports suggest that high environmental or occupational exposures to the fuel oxygenate methyl *tert*-butyl ether (MTBE) may result in breath concentrations that are sufficiently elevated to cause a false positive on commercial breath-alcohol analyzers. We evaluated this possibility in vitro by establishing a response curve for simulated breath containing MTBE in ethanol. Two types of breath-alcohol analyzers were evaluated. One analyzer's principle of operation involves in situ wet chemistry (oxidation of ethanol in a potassium dichromate solution) and absorption of visible light. The second instrument uses a combination of infrared absorption and an electrochemical sensor. Both types of instruments are currently used, although the former method represents older technology while the latter method represents newer technology.

The percent blood alcohol response curve was evaluated over a breath concentration range thought to be relevant to high-level environmental or occupational exposure (0–361 μg/l). Results indicate that MTBE positively biases the response of the older technology Breathalyzer™ when evaluated as a single constituent or in combination with ethanol. We conclude that a false positive is possible on this instrument if the MTBE exposure is very high, recent with respect to testing, and occurs in combination with ethanol consumption. The interference can be identified on the older technology instrument by a time dependent post-reading increase in the instrument response that does not occur for ethanol alone. In contrast, the newer technology instrument using infrared and electrochemical detectors did not respond to MTBE at lower levels (0–36 μg/l), and at higher levels (>72 μg/l) the instrument indicated an "interference" or "error". For this instrument, a false positive does not occur even at high MTBE levels in the presence of ethanol. © 2001 Elsevier Science Ireland Ltd. All rights reserved.

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1. Introduction

The partitioning of volatile organic compounds (VOCs) into and out of the blood provides convenient biological access for environmental and occupational exposure monitoring as well as evidential blood-alcohol testing [1–3]. The current study stems from questions raised at the intersection of these applications. Does the environmental contaminant, MTBE, interfere in the analysis of breath for blood alcohol? This specific question was raised by New Jersey state officials during the summer of 1995 when they were asked to respond to a claim that MTBE caused a false positive on a breath-alcohol test after occupational exposure to gasoline oxygenated with MTBE [4]. This claim appeared plausible since: (1) MTBE use as a gasoline oxygenate additive had

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been recently expanded as a consequence of the Clean Air Act Amendments of 1990; (2) the breath-alcohol analyzers used in New Jersey are an older technology (1954) based on nonspecific visible light absorbance; (3) recent studies had indicated that MTBE exposures can be high; (4) MTBE has a relatively low blood/breath partition coefficient, i.e. it readily partitions out of the blood into the breath; and (5) MTBE exposure and Breathalyzer™ testing are linked in time and space by automobile use activities [5–10].

For as long as evidential breath-alcohol analyzers have been used, there has been concern over interferences that could lead to false positive determinations [11]. Suspect VOC interferences include acetone, methane, and isoprene which are produced endogenously and can appear in the breath in high concentrations [12,13].

The ability of breath-alcohol analyzers to identify interferences varies with the principle of operation of the instrument's detector. Newer technology analyzers rely on IR absorption at multiple wavelengths (e.g. 3.39 and 3.48 μm) or use multiple detectors to either eliminate or identify interferences. Jones et al. [14] recently showed that a multiple wavelength instrument correctly identified interference from high levels of blood acetone, isopropanol, and/or methyl ethyl ketone. The question of an MTBE interference from the case in New Jersey relates to an older technology breath-alcohol analyzer — the Breathalyzer™ 900 (National Draeger, Inc., Durango, CO) — that is still used in that state as well as in New York. According to a survey conducted by the state of Colorado in 1995, there were 797 and 920 instruments being used and 30,000 and 11,300 tests conducted in each of these respective states [15]. The principle of operation for this instrument is based on the in situ oxidation of ethanol in a potassium dichromate solution and absorption of visible light after a fixed time of development. Since this technology relies on a single detector using visible light absorption, its specificity is based on ethanol's time dependent colorimetric reaction with potassium dichromate in a sulfuric acid solution. The specificity of this instrument has been established by ruling out interference from suspect VOCs [16]. Dubowski and Essary demonstrated that acetone does not present an interference on the Breathalyzer™ up to 2.9 μg/l [17].

In order to contrast the less specific detection of the older technology Breathalyzer™ to a current technology multi-detector instrument, we evaluated the Breathalyzer™ in parallel to the Alcotest™ 7110 (National Draeger, Inc., Durango, CO). This instrument uses dual detector technology including infrared absorption at 9.5 μm and electrochemical detection. Both types of instruments are classified as quantitative evidential analyzers meaning that the blood alcohol measurement provides sufficient evidence for establishing legal intoxication.

Exposure to volatile organic components of gasoline including MTBE is unavoidable in the environment and for some occupations, such as auto repair and gas station attendants [18]. As with other gasoline hydrocarbons, such as toluene, benzene, and xylenes, exposure to MTBE can occur through inhalation, ingestion, or percutaneous routes. MTBE is added to fuel to enhance octane and as an oxygenate to comply with the 1990 Clean Air Act Amendments for control of carbon monoxide and ozone [19]. It is estimated that oxygenates are currently added to one third of the nation's fuel used by 30 million people in this country. In 1996, 10 billion kg of MTBE was used in US gasoline which was an increase of 50% over the year before [20].

Even though exposure to ethanol is much greater for intentional ingestion relative to that occurring due to trace level VOC contamination of air or water, breath concentrations of MTBE and ethanol may be comparable since ethanol has a blood breath partition coefficient ($\lambda_{blood/breath}$) 100-fold greater than that of MTBE. Relative to ethanol, MTBE and other VOCs partition from the blood much more readily. Based on in vitro and human in vivo techniques, Nihlén et al. and Buckley et al., respectively, estimated MTBE's $\lambda_{blood/breath}$ to be 18 [9,10]. In contrast, the accepted blood breath partition coefficient for ethanol is 2100 [21]. This means that at equilibrium, for the same concentration in blood, the concentration of MTBE in breath will exceed ethanol by over 100-fold, or in other words, the concentration of MTBE in blood can be 100 times less than ethanol and the breath concentrations will be the same.

One of MTBE's primary metabolites, *tert*-butyl alcohol (TBA), could also potentially present an interference on the breath-alcohol analyzer. The potential for interference among MTBE and TBA will depend on their relative concentration in blood and their relative blood breath partition coefficient. Data regarding their relative concentration in blood varies. In a controlled exposure experiment, Buckley et al. showed blood MTBE concentrations exceeded TBA [9], while the opposite relationship was observed by Moolenaar et al. [22] for 12 occupationally exposed workers. Results from this study indicated that median values of TBA exceeded MTBE by a factor of 4 and 12 during two different winter phases [22]. With respect to the $\lambda_{blood/breath}$, MTBE is strongly favored for the potential for interference based on a $\lambda_{blood/breath}$ 27 times lower than TBA (18 versus 462) [10].

The goal of the current study was to evaluate whether MTBE breath concentrations present an interference on commercial evidential alcohol breath analyzers. This study was conducted to address inquiries by state and local officials receiving claims of such an interference associated with occupational exposure. This research was deemed to be important because in most states, Breathalyzer™ results alone are sufficient to establish legal intoxication. Therefore, a false positive reading can potentially lead to a conviction carrying profound financial, psychological, and social consequences. Furthermore, such claims, if left unsubstantiated, potentially jeopardize public acceptance of the oxyfuel program.

2. Methods

Two Breathalysers were evaluated: the Breathalyser™ 900A and the Alcotest™ 7110 (National Draeger, Inc.,

Durango, CO) each representing a different generation of technology and principle of detection. The Breathalyzer™ 900A instrument was developed in the 1950s and its detector is based on visible light absorption. The Alcotest™ 7110 device represents current technology and it has two independent detectors each with a different principle of operation. The first detector is based on infrared spectroscopy at 9.5 μm (rather than 3.4 μm of previous technologies where organic interferences are more likely). The second detector is electrochemical.

The breath alcohol instruments were evaluated using simulated breath made up of humidified zero air. Humidified zero air is a reasonable surrogate matrix for breath since both are made up primarily of nitrogen (79% versus 76%), oxygen (21% versus 15%), and water (2.6% in both cases). It is unlikely that the remaining minor constituents, such as carbon dioxide or argon have a matrix effect since the accepted legal method for calibrating Breathalyzers relies on air purged through a standard premixed water/ethanol solution designed to yield a specific ethanol concentration.

Simulated breath samples were prepared in pressurized 6 L Silcosteel™ canisters (Restek Corp., Bellefonte, PA) from neat solutions using well established procedures [25]. Samples were prepared by injecting a 200 μl aqueous solution containing 190 Proof Punctilious™ ethanol (Quantum Chemical Corp., Cincinnati, OH) (37 mg ethanol/ml water) and varying amounts of liquid MTBE (99.8% HPLC grade, Aldrich Chemical Co., Milwaukee, WI) to yield the desired concentration in the canisters when pressurized to 4× ambient with zero grade air. At standard temperature and pressure, the resulting humid air sample contained 312 μg/l ethanol, and 0–361 μg/l of MTBE. The exact volume of air added to each canister was determined by differential pressure measurement using an Absolute Pressure 1500 Bourdon gauge (Wallace & Tiernan, Belleville, NJ) of the evacuated and pressurized canisters at room temperature. The 312 μg/l ethanol (molecular weight of 46.07 g/mol) corresponds to 0.064% blood-alcohol according to Dubowski who showed that percent blood alcohol is given by gram ethanol/210 l air at 34°C [3]. This conversion assumes a blood–breath partition coefficient of 2100 which is the accepted value for the industry [21].

Laboratory personnel were trained to operate the blood alcohol test instruments according to manufacturer's specifications which are described here in brief. The Breathalyzer™ 900A instrument was allowed to warm-up for a period of 30 min so that the temperature of the sample chamber was within the range of 47–53°C. Prior to sample analysis, the instrument was zeroed with ultra pure zero air. Samples and zero air were analyzed by identical procedures. The pressurized samples were introduced into the analyzer via Teflon tubing and a needle valve at a flow rate of 2 l/min into the 52.5 cubic centimeter (cc) cylinder contained within the analyzer. Once the cylinder was filled, a green light was lighted prompting the operator to close the "take" valve and activate a plunger within the cylinder which pushes the collected sample through a second valve where it was bubbled into a disposable ampule containing a solution of potassium dichromate and sulfuric acid. Before use, the ampule diameter and solution level was checked by a gauge provided by the manufacturer. The solution was allowed to develop for about 90 s after which time the operator was prompted by a second indicator light to activate the tungsten detector lamp. Once the lamp was activated, the instrument's photometer measures light absorbance in the sample ampule relative to a blank ampule. Percent blood-alcohol was read by the deflection of a pointer caused by balancing the instrument's galvanometer. A record of the reading was provided by an ink imprint of the needle pointer on a test ticket. Additional readings were made at 30 s intervals for 5.5 min in order to assess whether MTBE's colorimetric reaction might differ from ethanol's thereby providing a means for identifying the interference.

The second instrument representing current technology was a Draeger Alcotest™ 7110. In contrast to the 900A, this instrument was microprocessor controlled, fully automated, and programmable. The simulated breath sample was passed through the instrument's 46 in. heated breath hose into 70 cm^3 cuvette. The instrument was factory programed to accept a specified rate and volume of sample. The sample, room air, and a calibration check gas was introduced to the analyzer through a preselected program. For each sample analysis, the following sequence was selected: (1) blank; (2) calibration check; (3) blank; (4) sample; (5) blank; (6) sample; (7) blank; (8) calibration check; and (9) blank. Therefore, each sample was run in duplicate, bracketed by calibration checks, and interspersed with five blank analyses. For each run, the results from both the infrared and fuel cell detectors were provided along with a time stamp, the breath volume (in liters) and the "blow time" (in seconds).

Both instruments were factory-calibrated. The instrument's calibration was checked using commercially available Certified Simulator Solution (Plus 4 Engineering, Eagle Vail, CO) prepared to yield a breath alcohol reading of 0.100% blood alcohol at 34°C. At the beginning and end of each day's run of analyses, the calibration of both alcohol breath analyzers were checked by a device known as a "Simulator" designed for such purposes. When a commercially available certified standard ethanol solution was added to the simulator, it was designed to generate a head space ethanol concentration corresponding to 0.10% blood alcohol at 34°C. This head space air was then pumped or drawn into the analyzer. The analyzer was considered to be within calibration if it reads within ±10% of the certified value.

After testing in each instrument, samples were analyzed qualitatively by an ITS40 ion trap GC/MS (Finnigan MAT, San Jose, CA) using a Nutech 3550A cryofocusser (Graseby Andersen, Smyrna, GA) to confirm the purity of the MTBE, water, and ethanol components of the sample.

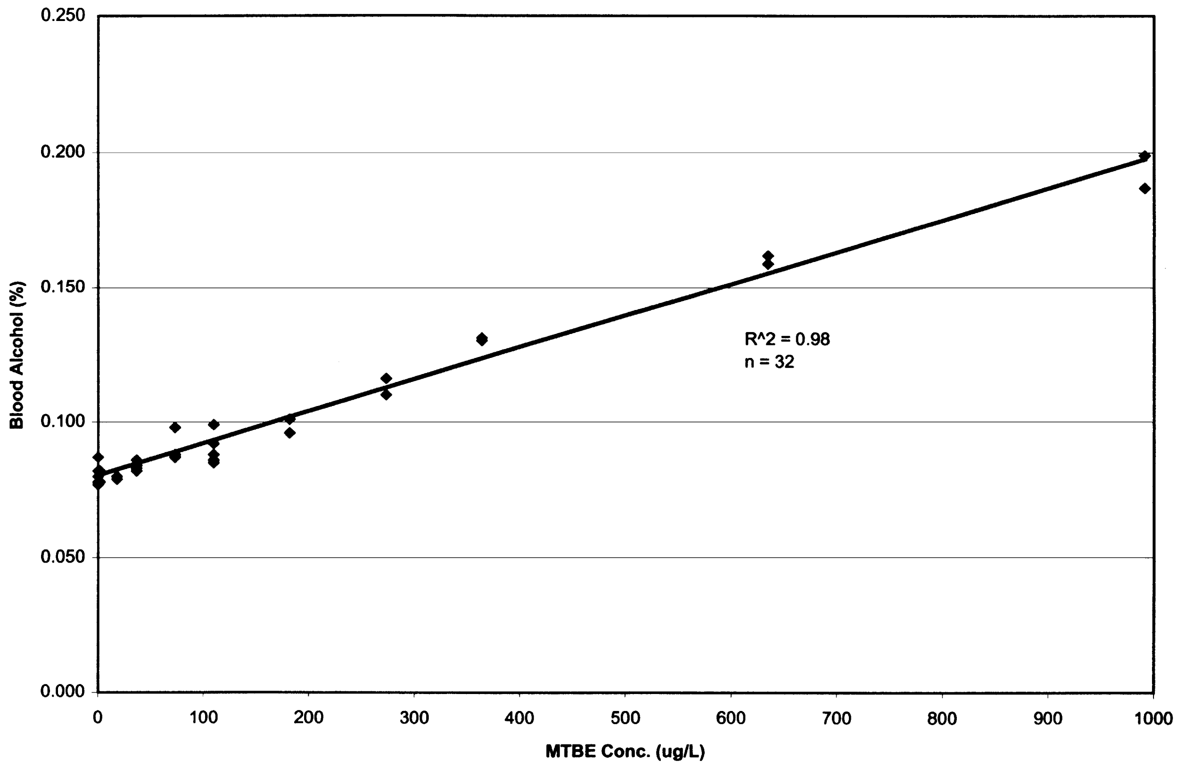

Fig. 1. Response of Breathalyzer™ 900A to MTBE in simulated breath with 324 μg/l ethanol.

3. Results

The response of the Breathalyzer™ 900A to simulated breath samples containing 100% relative humidity, a constant ethanol concentration (312 μg/l) and concentrations of MTBE that varied from 0 to 991 μg/l is shown in Fig. 1. The Breathalyzer™ response was recorded after the 90 s development period indicated by the instrument. Although the lower concentrations of MTBE in breath (i.e. <363 μg/l) are of greatest interest because of their environmental and/or occupational relevance, the values of 635 and 991 μg/l have been included to show the consistency of the linear relationship across a wide concentration range. For both the concentration range 0–363 μg/l ($n = 28$) and 0–991 μg/l ($n = 32$), regression analysis yielded a significant model ($P < 0.0001$) with $R^2 = 0.91$ and 0.98, respectively. The effect of repeated blood alcohol measurements (for $n = 32$) was taken into account using SAS's "Proc Mixed" procedure which gave estimates of the slope and intercept (standard errors are shown in [·]) as shown in Eq. (1).

$$\%BL_{EtOH} = 0.000429[0.000015](C_{MTBE}) + 0.081[0.0017] \quad (1)$$

MTBE was tested in combination with ethanol because initial tests with MTBE alone (2–692 μg/l) yielded a significant ($P < 0.001$) linear regression with a slope (0.00042) too small to result in a false positive for breath concentrations within a realistic range. With this slope, a breath concentration of 865 μg/l would be needed to achieve a percent blood alcohol of 0.10 on the Breathalyzer™ 900A. It is important to note the consistency between the slopes for the regressions (MTBE alone and MTBE with ethanol) suggesting that the Breathalyzer's MTBE response is independent of the ethanol concentration.

A screening assessment of TBA's interference was conducted to verify that MTBE posed the primary interference rather than its metabolite. The Breathalyzer™ response was assessed for MTBE and TBA at two levels (321 and 803 μg/l) in 312 μg/l in ethanol. At each level, the MTBE to TBA Breathalyzer™ response ratio was >1 (1.24 and 1.17, respectively) indicating that MTBE presented the greater interference. Considering TBA's lower response and greater blood breath partition coefficient, TBA was judged to present a negligible interference risk relative to MTBE and no further analysis of TBA was conducted.

Over the course of study with the 900A instrument, it was observed that when MTBE was present, the instrument response continued to increase whereas no increase was observed for ethanol alone. We decided to further characterize and evaluate this observation as a possible means for identifying an MTBE interference. Readings from the 900A instrument were collected at 30 s intervals for 5.5 min past the instrument's "read" indicator. The change in instrument response is shown by plotting the ratio of the reading at time

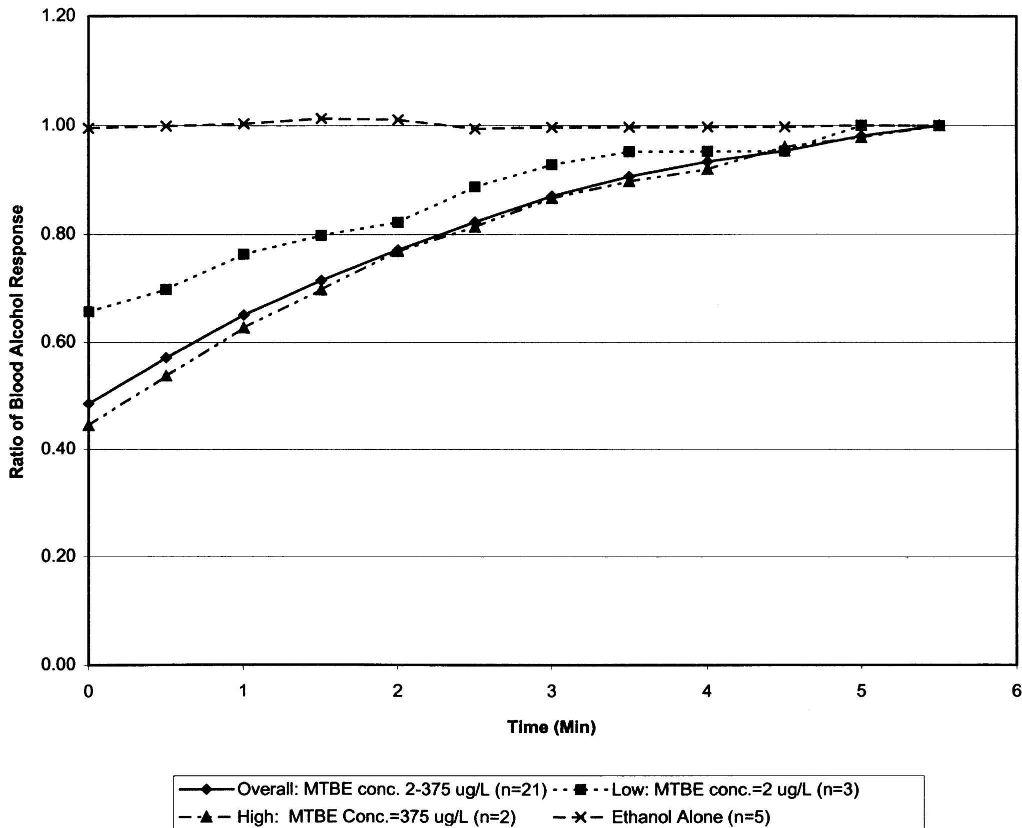

Fig. 2. Increase in Draeger™ 900A response at times following the instrument indicated initial reading (time = 0 min) relative to an unchanging time-course for ethanol when no MTBE was present.

0 (when the "read" indicator light was shown) to the reading at subsequent time points. The increasing ratio indicates that the instrument's response continued to increase. There appeared to be an MTBE concentration dependent curve profile such that the change over time was most pronounced for MTBE at high concentrations. The ratio remained constant when no MTBE was present Fig. 2.

Fig. 3 shows MTBE's response on the Draeger 7110 instrument which relies on infrared absorbance (9.5 μm) and electrochemical detection. Although the instrument gave a response in all but two cases (2.0 and 36 μg/l MTBE) the reading was accompanied by a "mouth alcohol" or "interference" flag invalidating the test.

4. Discussion and conclusions

The risk of a false positive on commercial breath analyzers is primarily dependent on two factors: the concentration of MTBE in breath and the sensitivity with which the analyzer responds. The current study has evaluated the Breathalyzer's™ response to MTBE in simulated breath. The relevant range of MTBE breath concentrations can be gleaned from the published literature where levels of exposure or body burden have been reported (see Table 1). For each study, the MTBE measurement representing the worst case or peak exposure was identified since sample sizes in general were small and because peak measurements would provide a conservative breath concentration estimate. Because there are very few published breath measurements, additional breath values have been estimated from air and blood concentrations since the relationship between these matrices and breath have been defined. Breath is estimated from a blood concentration by assuming a blood-breath partition coefficient of 18 as has been reported by Buckley et al.[9] and Nihlén et al. [10] using in vivo and in vitro techniques, respectively. The breath concentration can be estimated from an air concentration by f, the fraction of C_{air} exhaled unchanged in breath [24]. An estimate of f is given by Buckley et al. as 0.53 as the mean for two individuals from a controlled MTBE inhalation exposure [9].

Using a simulated breath matrix (100% humidification) prepared in summa polished canisters, it has been established that MTBE presents a positive bias in the measurement of percent blood alcohol on an older technology Breathalyzer™ relying on in situ chemical oxidation of

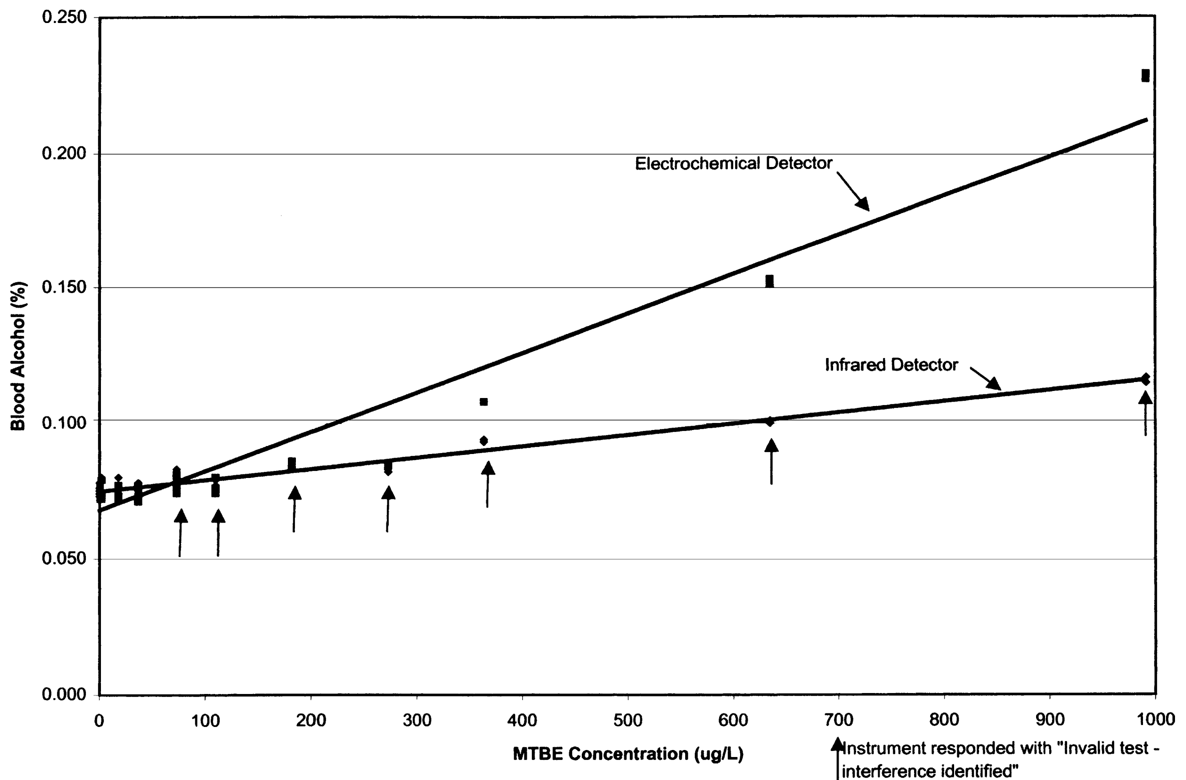

Fig. 3. Response of Draeger™ 7110 to MTBE in simulated breath with 324 μg/l ethanol.

ethanol and spectrophotometic detection. It may be possible to identify the MTBE interference by noting an increase in the instrument's percent blood alcohol reading after the initial reading. The same increase does not occur when ethanol alone is present. These results suggest that the colormetric reaction of MTBE and ethanol occur on a different time scale.

The newer technology instrument employing infrared and electrochemical detectors showed no bias and/or successfully identified the interference. These results are consistent with Josty, who observed that MTBE concentrations of 37 μg/l did not produce elevated readings on a similar infrared device [25]. In the older technology instrument, the bias was small, however, and only likely to result in a false positive under extraordinary occupational or environmental exposure circumstances and when this exposure occurs in addition to ethanol ingestion. This bias occurs across a concentration range which has been shown to be relevant for high-end short-term environmental exposures (refueling) or daily average occupational exposures. Extraordinary occupational exposures may result from a combination of inhalation and dermal exposures, such as might occur in an auto repair facility especially if hygiene practices are poor. Extraordinary environmental exposures might occur with contaminated drinking water creating the circumstances for combined exposure from ingestion (e.g. drinking water), inhalation (e.g. off gassing into the residential environment), and dermal absorption (e.g. from showering or bathing).

Factors favoring MTBE's positive bias include the fact that it is detected by the Breathalyzer™ with greater sensitivity than is ethanol and that MTBE more readily partitions from the blood into the breath (factor of 100). These factors are offset by the fact that exposure to ethanol is orders of magnitude greater than MTBE due to the intentional ingestion of ethanol.

Although it has been shown that MTBE in breath results in a positive interference on the breath analyzer, it is important to recognize that this interference is only substantial at breath concentrations that result from unusually high exposures. Furthermore, even if the exposure is substantial, a Breathalyzer™ false positive (with respect to the legal threshold of intoxication) is only likely to occur if ethanol has been ingested in addition to the exposure. To place this into perspective, of the approximately 100 persons monitored across the seven studies summarized in Table 1, the highest breath concentration of 7 μg/l estimated from personal monitoring while refueling would increase the Breathalyzer™ response by only 1%. Exposure to the ACGIH occupational threshold limit value of 144 μg/l would yield a 5% increase in the Breathalyzer's™ response.

It is concluded that MTBE does potentially present positive interference on the older version Breathalyzer™

Table 1
Actual and/or estimated MTBE breath concentrations from published environmental and occupational exposure studies

Breath concentration (μg/l)	Estimated from	Study	Reference
Actual breath measurements			
0.943		Chamber study, two subjects, the mean of two peak values is reported	[9]
0.810		Concentration in a single subject after refueling	[23]
Estimated from reported blood values[a]			
0.144	2.6 μg/l	Maximum venous blood concentration measured among 14 commuters in Stanford, CT	[8]
1.61	28.9 μg/l	Maximum concentration among three station attendants in Stanford, CT	
2.04	36.7 μg/l	Maximum concentration among 13 car repair workers in Stanford, CT	
0.10	37.0 μg/l	Maximum concentration among 18 workers in December 1992 in Fairbanks, AK when fuel oxygenate program was in place.	[22]
Estimated from air measurements[b]			
7.35	14700 μg/m^3, 4.1 ppm	Peak personal air concentration measured while refueling at a pump without Stage II controls	[6]
68.5	137000 μg/m^3, 38 ppm	Maximum integrated 2 min measurements while refueling without vapor recovery. Gasoline contained 12–13% MTBE.	[26]
6.99	13900 μg/m^3, 3.9 ppm	Peak personal air measurements ($n = 41$; 4 h integrated samples) among service station attendants in Phoenix where the average content of MTBE in gasoline ranged from 12.4 to 13.2%.	[7]
245	122500 μg/m^3, 34 ppm	Peak concentration measured on persons during refueling ($n = 313$)	[27]
126	63115 μg/m^3, 18 ppm	Highest measured occupational short term exposure levels (STEL) at a petroleum refinery and transport loading facility ($n = 38$)	[28]
Occupational air exposure guideline (ACGIH TLV)[b]			
72	144000 μg/m^3, 40 ppm	The time-weighted average concentration for a conventional 8 h workday and a 40 h workweek, to which nearly all workers may be repeatedly exposed, day after day, without adverse effect.	[29]

[a] The breath concentration is estimated from a blood measurement by assuming a blood breath partition coefficient of 18, i.e. C_{breath} (ng/l) $= C_{blood}$ (ng/l)/18.

[b] The breath concentration is estimated from an air measurement by assuming a f value of 0.5, i.e. $C_{breath} = C_{air} \times 0.5$. By definition, f is the fraction of C_{air} exhaled in breath unchanged at equilibrium [24].

relying in situ oxidation and spectrophotometric detection, however, a false positive is very unlikely unless extraordinary exposures have occurred and is in combination with ethanol ingestion. Newer technology Breathalyzer™ employing infrared and electrochemical detectors are not susceptible to this interference.

This research can be logically built upon by confirming the results of this study with Breathalyzer™ tests on individuals with high level occupational exposures to MTBE, such as auto mechanics.

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