Variables Affecting the Accuracy and Precision of Breath Alcohol Instruments Including the Intozilyzer 5000

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Breath testing instrument have been used extensively for the determination of ethanol in medicolegal investigations due to the simplicity in their operation, the relative portability of the instruments and the immediately available results. Unfortunately, breath alcohol instruments universally are prone to false positives (i.e. instrument reports ethanol is present when in fact it is responding to a different chemical) and falsely elevated breath values when ethanol is present (due to a variety of factors). The Intoxilyzer 5000 breath alcohol instrument is no exception and numerous studies have documented some of the sources of the errors with this instrument. The major types of variables discussed in this chapter include physiological (biological) variables and analytical (instrumental) variables with overlap between the two. The uncertainty in evidential breath-analyzer readings for a random subjects in the post-absorptive state has been determined to be as much as ±27% with over 90% of this uncertainty due to biological variables of the subject and at least 23% of subjects having their actual blood-alcohol concentration overestimated (Simpson 1987). The variables affecting the precision and accuracy of breath alcohol measurements discussed below are summarized in Table 1.

2100:1 Blood:Breath Partition Ratio

Many literature articles and breath test training manuals describe the partitioning of ethanol from the blood to the breath as being governed by Henry's Law. This law states that when a volatile chemical (alcohol) is dissolved in a liquid (blood) and is brought into contact with a closed air space (alveolar breath) an equilibrium is rapidly formed and there exists a fixed ratio between the concentration of the volatile compound in air and its concentration in the liquid at a given temperature. In **order to understand the application of this Law, one needs to imagine a capped bottle containing water and a little ethanol. The bottle will contain the water and ethanol in two forms; liquid, and gas above the liquid. This law states that at equilibrium, one can measure the concentration of the ethanol in the gas phase, and from that measurement predict the concentration in the simultaneous liquid phase. The comparison being made is that the lungs are like the bottle, the blood in the lungs are like the liquid in the bottle, and the breath is like the gas phase above the liquid. Unfortunately, Henry's Law does not apply in the lungs. In order for Henry's Law to apply, three conditions must be met. One, the solution must be in a closed system, like a sealed bottle. The lungs are open, not closed. Two, the solution must be kept at a known, constant temperature. The lung temperature is never known, and the temperature is always changing. And three, the pressure must be kept constant. The lungs are always changing pressure, decreasing pressure to inhale and increasing pressure to exhale.** Without all **three conditions present, it is not possible for equilibrium to occur, and Henry's Law does not apply.**

Table 1. Summary of representative studies of variables affecting the precision and accuracy of breath alcohol measurements and references.

Variable	Examples	Affects on Breath alcohol concentration	Ref.
Blood/ Breath Ratio	Observed Partition Ratio	Average 2215:1 ± 198 (Std. Dev.)	Dubowski 1985, Moore 1991, Jones 1993, Jones et. al. 1992, Sim son 1987
	Suggested Partition Ratio	1880:1 for <1% overestimation 1720:1	Thompson 1997 Dubowski 1985
Blood Water Content	Hematocrit fractions	6.4% uncertainty	Thompson 1997
Absorption Elimination/ Retrograde Extrapolation	Metabolism of ethanol	12+/-4 mg/h for non drinkers, 15+/-4 For social drinkers, and 30+/-m for alcoholics	Winek and Murphy 1984
	Absorptive state	230%,+190%, and +60% uncertainty	Simpson 1987
Breathing	Hyperventilation Breath holding	55% below the true BAC 14% above the true BAC	Ohlsson et. al. 1990
Body Temp.	Hyperthermia H thermia	8.62% increase per degree 6.8% decrease per degree	Fox and Hayward 1989 Fox and Ha and 1987
	Breath simulator temperature	6.25% increase/decrease per degree increase/decrease	Memari 1999
	Theoretical	6.5% per degree	Thompson 1997
Interfering Volatile Organic	Isopropanol	18.4 -123% increase in true BAC, 0.0116-0.0292, 0.011, 0.009 2101, increase	Logan et. a 1994, Cowan et. al. 1990, Dubowski 1991, Memari 1999
Compounds	Acetone	0.0116 g/210L increase, Interferent indicated	Cowan et. al. 1990, Memari 1999
(false	m-xylene, o-xylene	0.042, 0.034 2101, increase	Caldwell and Kim 1997
Positives)	Methyl ethyl ketone	0.0286, 0.004 g/2101, increase	Cowan et. al. 1990, Memari 1999
	Methanol	0.20, 0.395, 0.032 g/2101, increase	Cowan et. al. 1990, Caldwell and Kim 1997, Memari 1999
	Toluene	0.0294, 0.028 g/2101, increase	Cowan et. al. 1990, Memari 1999
	Acetaldehyde	0.01 2101, increase	Memari 1999
	Combination	0.071-0.083 2141, increase	Memari 1999
Systematic Errors (Bias)	i.e. standards calibrating out of the range	Instrument calibrates with solution out of range +/- 10% 25% of the time	Memari 1999
Gross Errors	Ethanol standard run with unknowns	Without positive controls reliability at test time unknown	
Random Errors	Confidence intervals must be calculated	If not calculated can lead to falsely elevated reported reading of +0.02 or eater	

At best, Henry's law can only be used as an approximation and a recent detailed derivation of the partition ratio using the simplest water-air-ethanol system present in "simulator solutions" concluded that if a fixed ratio is used it ought to be 1880:1 (Thomson, 1997). Previously, Dubowski has suggested an even lower fixed value of 1720:1 (Dubowski, 1985). Currently, the value of 2100:1 is routinely incorrectly used by manufacturers, law enforcement agencies and legislatures as a fixed partition able to accurately predict blood alcohol levels, when numerous studies throughout the years have shown that this is not the case and significant errors can occur when using a fixed 2100:1 ratio (Alobaid 1976, Jones 1983, Thompson 1997). The concept that ethanol easily evaporates from the pulmonary circulation and diffuses into the alveolar airspace to be exhaled with each breath is the basis for believing the so-called blood-breath partitioning of ethanol at a fixed ratio. Recent research has demonstrated that ethanol does not diffuse from the pulmonary circulation to the alveolar air space as has been previously thought (Hlastala 1998). In fact the diffusion occurs from the pulmonary circulation to the conducting airways of the lungs, bypassing the alveoli. The significance of this work is that no stable partition ratio can exist for any given individual and certainly no partition ration can be predicted for any given person. As a result, the 2100:1 ratio will be incorrect most of the time.

Another problem with using the 2100:1 ratio is that law enforcement assumes the subject is in the elimination phase, that is, post-absorbed. However that information is usually lacking. The significance is that during the absorption phase (when ethanol is being absorbed into the bloodstream from the small intestine) the partition ratio is going to be lower compared to when absorption is complete. Therefore in every individual their partition ratio will change as the ethanol is absorbed, then eliminated. Generally it is not known whether a person is absorbing or eliminating ethanol at the time of their breath test. Additionally, the hematocrit and fraction of water in an individual's blood is variable and changes the partition ratio resulting in additional uncertainty (Thompson 1997). Also Martin et. al. **(1984) reported that during absorption, arterial BAC tended to be higher than venous BAC, peaking at a higher level (tmax) and with a shorter time to peak(tmax) until an arterao-venous concentration equilibrium was reached, whereafter VBAC remained above ABAC. Although there was a close relationship between BrAC, ABAC and VBAC during elimination, BrAC closely followed the pattern of ABAC during absorption and tended to deviate from VBAC. BrAC, therefore, is much better predictor of ABAC during absorption than** VBAC. **Simultaneous measurements of breath alcohol concentrations (BrAC) and venous blood alcohol concentration (VBAC) has shown that actual VBAC can be overestimated by more than 100% for a significant amount of time after drinking stops (Simpson 1987).**

Absorption/Metabolism of Ethanol and Retrograde Extrapolation

Difficulties with retrograde extrapolation are highlighted by considering the "Hip Flask" defense, whereby a motorist consumes a significant amount of alcohol beverage from their personal flask or elsewhere following a crash. There is no way of knowing what the real ethanol concentration was at the time of the crash if the usual one or two hours pass from the crash until the blood or breath test. This argument is logically extended to if a person has enough alcohol in his or her stomach at the time of a stop or crash, they will continue to absorb the ethanol for that time interval until the blood or breath test is given. The importance of this argument is that a person can absorb a lot of ethanol in a short time, up to 0.0025 gm/dl per minute, or 0.15 gm/dl per hour. So even if a person has a 0.20 gm/dl breath or blood test an hour after a crash, they

could have been an 0.05gm/dl at the time of the crash. Sometimes this extrapolation argument is used by law enforcement in the opposite way to extrapolate an even higher estimate of BrAC based on the assumption than the person is in the fully postabsorptive state and eliminating ethanol at a constant rate. Obviously it is impossible to know what state of absorption an individual is and the high degree of variability in elimination rates makes any type of retrograde extrapolation mostly guesswork unsuitable for scientific evidence in court. Studies have reported that evidentiary BrAC analyses performed within 2h of driving can provide reasonable estimates of the BrAC existing at the time of driving but than extrapolation is unwarranted (Gullberg and McElroy 1992 and Gullberg 1991). Simpson (1987) has reported that reanalysis of BrAC data indicate 68% of the population had their actual BAC underestimated, 16% were acceptably close to the actual BAC, and 16% were overestimated in the fully postabsorptive state. Therefore, breath test results may tend to overestimate actual BAC for significant amounts of time even after the peak BAC has been reached.

Watkins and Adler (1993) reported that the average time required to reach maximum BrAC was 41 min for both empty (after 6hr fasting) and full stomach conditions. The average elimination rate of ethanol was found to be significantly lower after meal (0.017 BrAC/h compared to 0.02 BrAC/h) but the time required to reach zero BrAC was not significantly different (5.01 h full stomach, 5.05 h empty stomach). Rogers et. al. **(1987) have reported that carbohydrate caused a significant increase in rate of metabolism of ethanol and fat or protein caused small but non-significant decreases. Winek and Esposito (1985) also reported that the absorption of alcohol is influenced by gastrointestinal contents, motility, and the composition and quantity of the alcoholic beverage. The vascularity of tissues influences the distribution of alcohol and their water content will determine the amount of alcohol present after equilibrium. Winek and Murphy (1984) reported that ethanol elimination is a zero order process. The mean ethanol elimination rate for non-drinkers of 12+/- 4mg/h and for social drinkers l5+/-4mg%/h, and for alcoholics rate of 30+/-9Yng%/h. A report by Cole-Harding and Wilson (1987) showed that women metabolized ethanol faster than men, there was a small gender difference in peak BAL and no gender difference in time to peak BAL. Dubowski (1985) reported that since alcohol pharmacokinetics parameters are subject to wide intersubject variability such as time to peak after end of drinking, rate of elimination, sex, and age, these variables make the blood alcohol information widely distributed and inappropriate as a guide for the drinking behavior of individuals.**

Breathing (volume, hyperventilation, breath holding)

The rate of breathing affects the concentration of ethanol in the breath. Hyperventilation causes a lower breath result (up to 55%) while hypoventilation (breath holding) increases the breath result (up to 14% higher than actual BAC) (Ohlsson et. al. 1990). **Both breathing disorders can be caused by disease, trauma, and drugs and should be considered as a source of potential error in breath testing. Others have reported that on a breath alcohol profile the area under the profile curve for samples preceded by breath holding is significantly larger than when breathing is normal prior to sample provision (Gullberg 1989). It has also been reported that exhaled air at the end of maximal expiration does not always provide the best, or a close, indication of the plasma(or blood) ethanol concentration particularly when conditions exist such as chronic obstructive pulmonary disease (Russell and Jones 1983).**

Body/Breath Temperature

Since heat is a driving force which causes the ethanol to diffuse from the blood to the breath, the higher the temperature the greater the amount of ethanol which will diffuse into the breath (keeping the blood concentration constant). A temperature of 34°C as been reported as an average temperature for end-expired breath in healthy men (Jones 1982 and Dubowski 1975). However, not all investigators agree (Hlastala 1998). An 8.62% increase for each degree C increase in core body temperature and 6.8% decrease per degree C in core body temperature has been reported (Fox and Hayward 1989, Fox and Hayward 1987). The apparent breath alcohol concentration using the simulator increases linearly with temperature with a 6.25% per degree C increase as seen in Figure 1 (Memari 1999). This could explain why the Intoxilyzer 5000 manufacturer requires higher concentrations for ethanol reference solutions to give proper readings at 34°C. For example, the manufacturer specified target value for a calibrator solution is 0.0968 to produce a 0.080 on the instrument presumed to be operating at 34 °C as apposed to normal body temperature of 37°C. Thus a four-degree rise in temperature will give a substantial 25% increase in apparent breath alcohol reading result. An individual's normal body temperature can vary several degrees centigrade in a given 24-hour period, with the lowest temperature in the early morning rising to its peak in the evening. The inter-individual body temperature can also vary a few degrees centigrade. Finally, body temperature can vary four or five degrees centigrade when a person is sick.

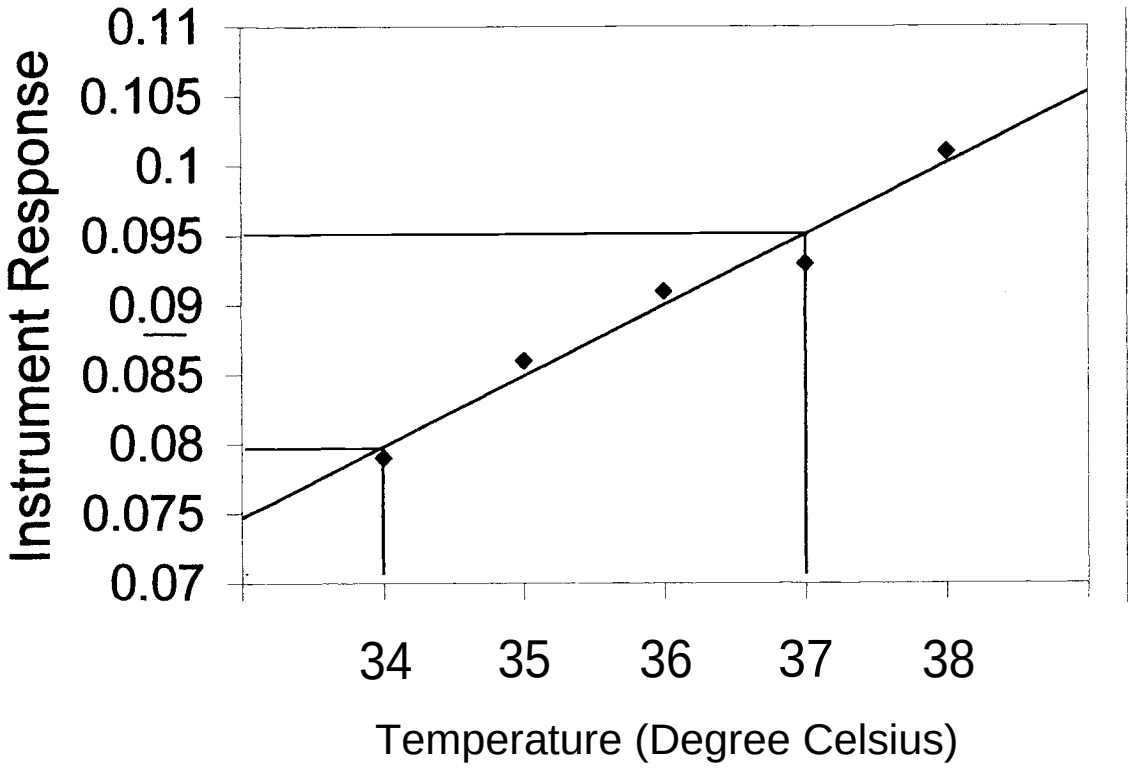

Figure 1: **Intoxilyzer 5000 Response to 0.0968 g/dL Ethanol Solution at Different Temperatures**

Interfering Volatile Organic Compounds

The Intoxilyzer 5000 is based on the transmittance of infrared energy at 3.80, 3.48 and 3.39 gm. Substances which also absorb infrared energy at similar wavelengths as ethanol will cause interferences. Human expired air consists of a mixture of gases including mainly oxygen, nitrogen, carbon dioxide, water vapor, and, in small amounts, a multitude of volatile organic compounds (VOCs). The VOC's expelled in breath are produced endogenously during normal metabolic activity or can be imbibed with food, drinks, inhaled from the ambient air in which they exist as atmospheric pollutants or result from occupational exposure. A summary of potentially interfering compounds at levels reported antimortem in human breath recently tested on the Intoxilyzer 5000 is given in Table 2 (Memari 1999). Compounds which were not found to significantly interfere were acetone, acetonitrile, isoprene, methyl ethyl ketone, trichloroethane and trichloroethylene. Compounds which produced an interference alert on the instrument were acetone and methylene chloride. At low levels isopropanol gave small false readings but at elevated amounts the instrument indicated interferrent present and stopped the test. Therefore, these compounds are unlikely to cause false positives with the Intoxilizer 5000 by themselves. Acetaldehyde yielded an apparent breath alcohol reading of 0.009 on calibration mode and 0.01 g/210L on sampling mode and does not activate the interference indicator meaning that the instrument cannot distinguish between ethanol and acetaldehyde. The two compounds tested which yielded the highest false ethanol responses were toluene and methanol producing apparent ethanol concentrations of 0.028 and 0.36 respectively. Methanol finds widespread commercial use as a solvent, especially in paints and varnishes. It is also a constituent of some antifreeze solutions, is used to denature ethanol, and is being considered as an alternative energy source. Methanol, in sufficient quantity, will produce a positive apparent alcohol concentration on the Intoxilyzer 5000, but will not illuminate an interference light.

Table 2. Response of Intoxil zer 5000 to Volatile Organic Compounds Found in Human Breath.

Concentration g/L solution	Apparent BrAC on Calibration mode 210L	Apparent BrAC on Sampling mode 210L
0.0044 g/L Acetonitrile	0.000	0.000
0.011 g/L Isoprene	0.000	0.000
0.10 g trichloroethane	0.000	0.000
0.050 g Methyl ethyl ketone	0.000	0.000
0.40g/L acetone	Interferent	Interferent
0.06g Methylene chloride	0.000	0.00(Interf. Sub.)
0.605 Ethanol	0.050	0.050
0.605g Ethanol + 0.068 Trichloroethylene	0.052	0.048
1.5 Isopropanol	0.047(Interf sub.)	Interferent Subtracted
0.26g Isopropanol	0.009	0.000
0.02g Acetaldehyde	0.010	0.008
0.605g Ethanol+0.0171g toluene	0.075	0.078
0.605 Ethanol + 0.02g toluene	0.083	Invalid sample
0.4g Methanol	0.032	0.036
0.40g Methanol+0.013g toluene+0.26g isopropanol+0.011 g acetaldehyde	0.083	0.071

Toluene is an aromatic petroleum hydrocarbon that has many important commercial and industrial applications as a solvent and starting material for organic syntheses. It is present in numerous paints, paint thinners, glues, and other products likely to be found in the household. It can be and has been, abused by individuals who inhale its vapor. Garriott considers the concentration "commonly found in abuse" for toluene to be about 1 to 30 mg/L (mean, 10 mg/L) in blood (Cowan et. al. 1990). Toluene alone can account for between 0.028 and 0.033 g/2101, of the ostensible ethanol reading without causing the interference mechanism to trigger (Table 2). However, if the signal resulting from toluene is augmented by the presence of ethanol, the readout could exceed legal limits without activating the interference light. Additionally, combinations of organic compounds can yield additive effects with four such compounds yielding apparent ethanol levels of 0.071-0.083 g/210L with no interference alert and no ethanol present as seen in Table 2. It is possible for such combinations to be present in individuals including due to environmental exposure or due to occupational exposure such as in the case of painters, chemists, etc. The reason this instrument also responds to these chemicals is that they absorb infrared radiation in the same region as ethanol as **seen on Figure 2 showing the infrared spectrum for ethanol compared to a combination of methanol and toluene with the wavelengths were the Intoxilyzer is reported to take measurement marked with vertical lines.**

The Intoxilyzer 5000 instrument has a "Custom Breath Test Mode Sequence With Sample Capture" which allows for the breath sample to be preserved on a tube containing a sorbent enabling one to reanalyze the sample at a later date (CMI 1989). Goldberger et. al. **(1987) have reported statistical analyses revealing good accuracy and precision and correlation between direct and delayed vapor ethanol analyses for instruments including the Intoxilyzer 5000 (range = 0.000 to 0.250g/210L, N=42/instrument, r greater than 0.99). Goldberger et.** al. **(1986) have also reported that collected breath samples on silica gel after retaining 1.25 to 2.75 years revealed good overall correlation between direct and delayed ethanol determination (r=0.900). Employing this quality control ensurance allows for the confirmation of breath alcohol readings as well as checking for any potential interfering organic compounds. Unfortunately this quality control measure is not universally employed and without such a sample it is impossible to know whether an Intoxilyzer reading was due to ethanol alone or due to combinations of the many other compounds which produce false breath alcohol readings.**

Analytical Errors

All measurements are subject to three types of experimental errors which need to be evaluated. The first errors are systematic errors which result from instrumental, method or personal errors and result in a bias of values positive or negative relative to the true value. Detection of this type of error is by the analysis of standard samples such as a calibration standard at the legal limit for the jurisdiction (i.e. 0.08g/210L) and correcting for any bias. The Intoxilyzer 5000 instrument has a "Control Breath Test Mode Sequence" which allows for just such a test (CMI 1989) but this quality control ensurance is not universally employed. The second errors are gross errors which may be rejected based on the widely used Q test for outliers. If a calculated Q_{exp} **is greater than** $Q_{f\,t}$ **taken from a reference table for a given number of measurements and confidence then a questionable result can be rejected.** Q_{eXp} **is difference between the questionable result and its nearest neighbor divided by the spread of the entire data set. The third errors are random errors which cannot be eliminated but are accounted for by determining confidence intervals. The magnitude of random errors are determined by the**

precision or repeatability and reproducibility. The exact value of the population mean, or the true value, g, from an analytical method can never be determined exactly because this would require an infinite number of measurements. Instead, a sample mean, ~, and a confidence interval is calculated using the statistical parameter t to produce confidence limits (Skoog et. al. 1994). Confidence limits define an interval around the sample mean, 4, which probably contains the population mean at a given confidence limit (ASTM E177-90a 1999). The true value, p = ~ ± ts/(N)ln. Where t is the t value from a reference table, s is the standard deviation of the data and N is the number of measurements. Confidence intervals are required for any analytical measurements including breath alcohol measurements.

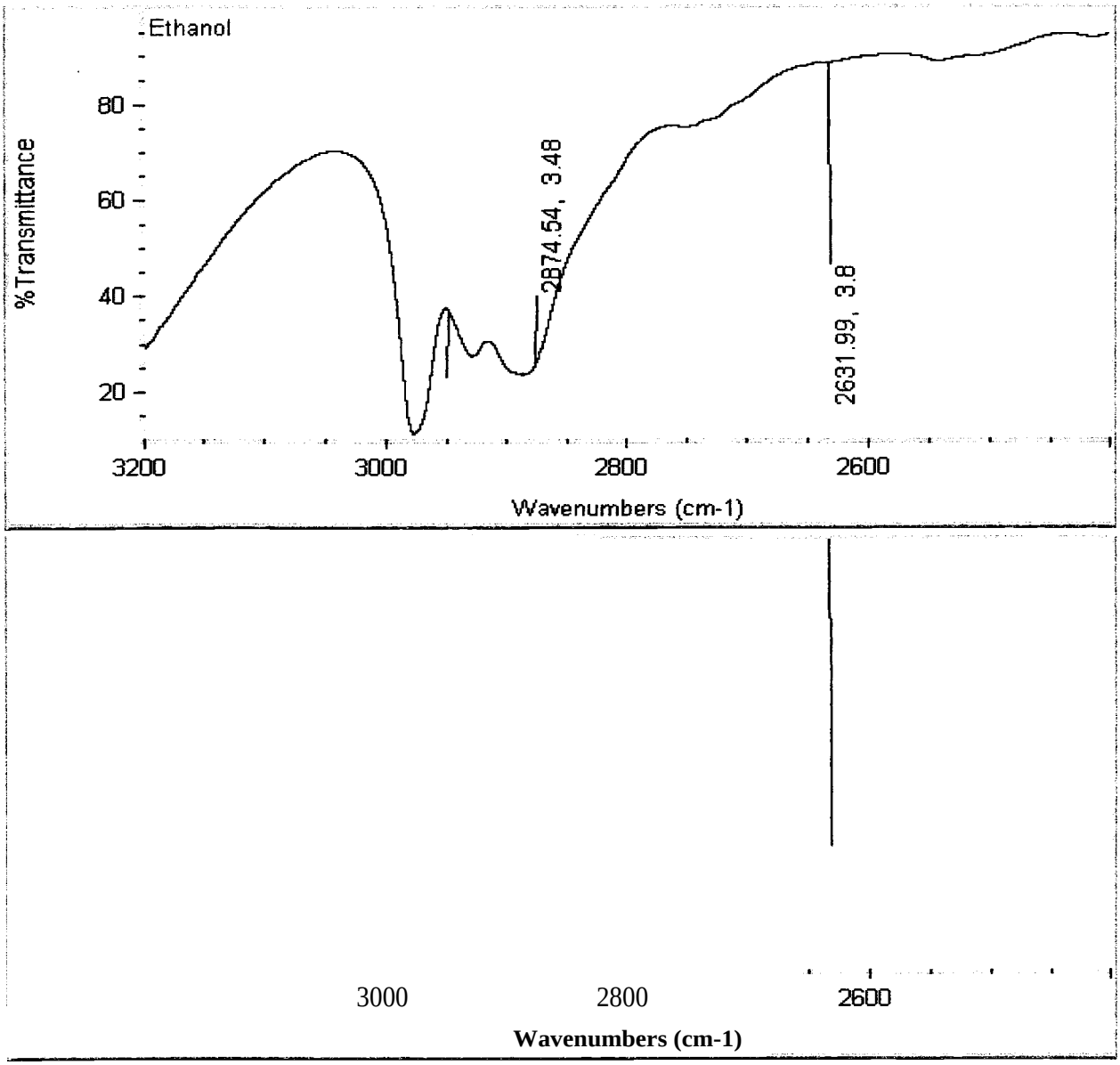

Figure 2. The FTIR Spectrum of Ethanol (top spectra) and a Combination of Methanol and Toluene (bottom spectra) in Vapor Phase Between 2400 to 3200 Wavenumbers (cm-1).

For example, the statistically significant confidence interval for widely varying hypothetical triplicate breath alcohol readings of 0.039, 0.060, 0.080 would be calculated as follows. First we must determine if any value can or should be rejected. $Q_{e,p} =$ **(0.060-0.039)/0.080-0.039) = 0.512. Q~rit for this data set is 0.994 for this data set at 95% confidence therefore, based on statistics we must retain the outlier. The true value,** $\sim_t = 4\ t\ ts/(N)'^2 =$ **(4.30)(0.0205)/[(3)"z] = 0.0597 ± 0.0508 at 95% confidence. In other words, the error in the measurement is nearly as great as the average value. In most cases, replicates are much closer than this extreme example and therefore the confidence interval would be smaller. In an attempt to account for some of the experimental inaccuracies of the Intoxilyzer 5000, duplicate measurements are required to be within ± 0.02% and highest value of the non-excluded data is generally used. This unfortunately is a completely scientifically unsound practice and is apparently used for convenience and to report the highest possible breath alcohol reading rather than the statistically valid number. Calculation of the average breath alcohol reading and the statistically sound confidence interval for such measurements is straightforward and could be easily incorporated into the breath alcohol instrument software or calculated in a few minutes with available data. The proper statistical processing and reporting of data is required in all areas of chemistry and most areas of forensic science excluding breath alcohol measurements. Proper scientific practice would include the reporting of the average measured value with confident intervals and preferably be reduced by a number to account for the high degree of uncertainty common in this type of measurement. Leading scientists including Dubowski and Jones have suggested that a correction factor ranging from 0.015 to 0.030 be subtracted from the mean of breath alcohol measurements (Labianca and Simpson, 1995) to correct for error from this source. In the absence of statistical analysis such a correction factor is the minimum that should be applied. Perhaps ironically, the most widely used introductory analytical chemistry textbook in the U.S. uses a triplicate blood alcohol determination as an example (Example 5-5) of statistical analysis with the values of 0.084, 0.089 and 0.079 yielding a true value, p = 0.084 ± 0.012% for 95% confidence (Skoog et al. 1994). And yet, this type of calculation is not routinely performed as it should be for breath alcohol measurements.**

Accuracy and Precision of the Intozilzer 5000 Using Calibrator Solutions

A recent study showed that with pure calibrator solutions under static conditions, the Intoxilyzer 5000 shows excellent calibration curve linearity and satisfactory precision as seen in Table 3 (Memari 1999). Relative standard deviations for ten measurements at the five calibrator solution concentrations was generally within 5%. This precision measure is a measure of the repeatability of the instrument with pure calibrator solutions under static conditions and separate from the error analysis discussed above required for actual breath sample analysis. Table 4 gives the manufacturer recommended reference solutions to yield breath readings of 0.05, 0.08, and 0.20 g /2101, by the Intoxilyzer 5000 and the reference solution allowed range by the Florida Department of Law Enforcement (FDLE). In order to measure the calibration error, solutions out of the allowed alcohol concentration range were prepared and analyzed on the Intoxilyzer 5000. Each ethanol solution was analyzed 15 times on calibration mode using the Intoxilyzer 5000. In this study, the Intoxilyzer 5000 calibrated with solutions outside the allowed range up to +/- 10% outside the allowed range 25% of the time (Memari 1999).

Table 3. Mean, standard deviation, and relative standard deviation for 10 measurements of calibrator solution on the Intoxilyzer 5000 instrument.

Reference Solution g/100ml	Target Vapor Concentration g/210L	Mean Intoxilizer Response	SD g/210L	RSD
0.025	0.020	0.020	0.001	6.2%
0.139	0.110	0.117	0.004	3.3%
0.254	0.210	0.212	0.005	2.4%
0.369	0.305	0.303	0.008	2.6%
0.486	0.400	0.397	0.019	4.7%

Table 4. Target values and allowed concentration ranges for ethanol reference solutions used to calibrate the Intoxilyzer 5000 instrument.

Breath Alcohol Cone. 210L)	Reference Solution Target Value fig/100mQ	Reference Solution Allowed range (g/100mL)
0.050	0.0605	0.0586 to 0.0623
0.080	0.0968	0.0938 to 0.0997
0.200	0.2420	0.2347 to 0.2492

Conclusions

While the Intoxilyzer 5000 demonstrates good analytical precision for standard solutions it suffers from numerous sources of error including calibration solution errors, temperature variations and interfering endogenous volatile organic compounds. These measured analytical instrument variations combined with the substantial biological variability (including absorption/distribution/elimination rate variations, blood/breath partition variations, etc.) warrant an extremely cautious approach to the reporting and use of breath alcohol readings using this instrument. These potential errors can and should be accounted for when using this instrument for evidentiary purposes and ultimately it should be used as a presumptive test with positive blood alcohol confirmation by dual column GC/FID or GC/MS analysis. Analytical reliability ensurances should always include body temperature (or instrument cell temperature) measurements (and include a correction factor), employment of the custom breath test mode sequence with sample capture for subsequent organic analysis of possible interferents and employment the control breath test mode sequence to ensure proper functioning of the instrument. Biological variables affecting reliability are more problematic and difficult to assess or correct for.

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