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**ORIGINAL ARTICLE**

# Monitoring people at risk of drinking by a rapid urinary ethyl glucuronide test

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**ABSTRACT**

Alcohol and illicit drug abuse are major public health problems worldwide. Since alcohol is the predominant substance of choice in polydrug abusers, monitoring its use, along with urinary drug screening in patients in rehabilitation programs, appeared to be crucial in identifying patients at risk of alcohol disorders leading to impaired quality of life. Ethyl β-D-6-glucuronide, a non-oxidative, non-volatile, stable and minor direct ethanol metabolite, has a 6h to 4 day window of detection in urine after the last alcohol intake. Each of the 119 subjects (85 males, 34 females) registered with the Public Health Service for Drug Dependence Treatment provided a urine sample for ethylglucoronide (EtG) determination in an immunochemical test with a 500 ng/ml cutoff. All results were evaluated with confirmation criteria of a fully validated gas chromatography/mass spectrometry assay. The diagnostic performance of the EtG immunochemical test was assessed using Receiver Operating Characteristic Curve analysis. The immunochemical test specificity was 100% for EtG urinary values above 500 ng/ml. No false positive results were found. With levels below 500 ng/ml, 12% of the samples were classified as negative. The average consumption of the incorrectly classified subjects was 171 ng/ml, with a misclassification error of 6.5% to 18.5%. High agreement between EtG as determined in an immunochemical test and gas chromatography/mass spectrometry, suggests that the rapid EtG test is a reliable, cost-effective alcohol monitoring assay for patient management in many non-forensic settings, such as drug rehabilitation programs.

**KEY WORDS:** ethyl glucuronide; alcohol biomarkers; ethyl glucoronide point of care test; urine analysis; gas chromatography/mass spectrometry

## Introduction

Polydrug use may include patterns of excessive drinking since alcohol is among the most frequently reported secondary substance problems for drug addicts (Gossop *et al.*, 2002). Some forms of drug misuse, *e.g.* cocaine, are closely associated with heavy drinking (Gossop *et al.*, 2006) and although opioid dependence is frequently associated with polysubstance use (usually cocaine, cannabis, amphetamines, benzodiazepines), alcohol is a major, if sometimes neglected, part of the overall pattern of multiple substance use (Gossop *et al.*, 2002; Soyka, 2015; Srivastava *et al.*, 2008). Even though methadone or buprenorphine maintenance therapy is established as first-line treatment for opioid dependence (Soyka, 2015), up to 40% of patients in opioid treatment programs were estimated to screen positive for an alcohol disorder. Indeed, alcohol abuse is often reported after drug addiction treatment (Gossop *et al.*, 2002).

Heavy drinking during drug rehabilitation programs deserves to be taken seriously as it causes multiple health and social problems (Hartzler *et al.*, 2010; Nyamathi *et al.*, 2009) and the risk of alcohol-related comorbidities (Nyamathi *et al.*, 2009; Klimas *et al.*, 2015). Drug and alcohol dependent clients had higher rates of criminal involvement, poor physical and mental health, including liver disorders, and worse prognosis when affected by chronic hepatitis C, social deterioration and increased risk of mortality (Klimas *et al.*, 2015; Staiger *et al.*, 2013; Best *et al.*, 1998; Roszell *et al.*, 1986). Alcohol abuse may also be a major factor in involuntary patient discharge from rehabilitation programs (Soyka, 2015). Consequently, monitoring

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alcohol use in patients in rehabilitation programs appears to be crucial for identifying those at risk of alcohol disorders, making brief interventions available to reduce unhealthy alcohol intake and, when necessary, referring patients to specialized treatment (Klimas *et al.*, 2015).

Although the golden standard for detecting recent alcohol use remains direct ethanol measurement in blood or breath, the short half-life of ethanol (several hours) decreases the sensitivity and utility of these methods in detecting alcohol relapses which occurred days previously (Leickly *et al.*, 2015). In clinical settings, standard biomarkers for diagnosing chronic alcohol abuse are included in the EDAC (Early Detection of Alcohol Consumption) panel, with the 12 most relevant routine tests being: sodium, potassium, chloride, total bilirubin, direct bilirubin, aspartate aminotransferase, gamma glutamyltransferase, HDL cholesterol, mean corpuscular volume, platelets, white blood cells and monocytes (Harasymiw *et al.*, 2005). The carbohydrate deficient transferrin (CDT) test, an iron transporter protein that is detected in serum as biomarker of heavy alcohol consumption, was recently added to the panel (Harasymiw & Bean, 2001). As its sensitivity is somewhat limited, especially in people with severe liver disease, CDT point-of-care analysis is not yet feasible (Leickly *et al.*, 2015; Bertholet *et al.*, 2014). None of these so-called "indirect markers" is sensitive or specific enough to determine the degree of alcohol abuse and its medical complications because all are influenced to some extent by factors like genetic variability, liver pathology, sex and age. For these reasons, attention has recently focussed on minor products of alcohol metabolism, the so-called "direct markers", which include ethyl β-D-6-glucuronide (EtG), ethyl sulfate, phosphatidylethanol and fatty acid ethyl esters. Resulting from non-oxidative ethanol metabolism, they are endowed with a wide detection window in urine (Kharbouche *et al.*, 2009; Maenhout *et al.*, 2013; Peterson, 2005).

EtG, a minor metabolite, forms in the liver through ethanol and glucuronic acid conjugation and can be collected in several body fluids, tissue and hair (Nanau & Neuman, 2015). Its urinary secretion accounts for approximately 0.02–0.06% of the ingested ethanol dose (Nanau & Neuman, 2015). It has greater specificity and sensitivity than all other known ethanol markers and is detected only after alcohol intake (Cabarcos *et al.*, 2015; Tarcomnicu *et al.*, 2010; Lamoureux *et al.*, 2009) for up to 36 h in blood and for up to 5 days in urine, which is the most feasible collection method at rehabilitation services (Leickly *et al.*, 2015).

EtG in urine is mainly determined by GC/MS (gas chromatography/mass spectrometry), liquid chromatography-mass spectrometry (LC/MS), or liquid chromatography tandem-mass spectrometry (LC-MS/MS). Although very sensitive, accurate and reasonably efficient, these technologies are costly and require highly skilled personnel (Jatlow & O'Malley, 2010).

A qualitative assessment, point-of-care (POCT) EtG immunochemical test is now available for on-site use. It can be conducted by non-technical staff, following a few simple steps and results are read within a matter of minutes, providing immediate feedback to the subject/patient (Leickly *et al.*, 2015). It generally uses the relatively conservative, standard EtG 500 ng/ml cut-off which was recommended by the Substance Abuse and Mental Health Services Administration (SAMHSA 2011), due to concerns about over-detection of alcohol use based on incidental non-beverage alcohol exposure, arising from ethanol ingestion in *e.g.* foods, mouthwashes, and other over-the-counter products. The recommended cut-off is also designed to discriminate between safe social drinking and heavy, hazardous drinking.

The aim of the present paper was to determine alcohol consumption by POCT EtG immunochemical test (EtG-I) as measured in the urine of 119 patients who were registered with the Public Health Service for Drug Dependence Treatment. Forty-eight patients were in maintenance therapy with methadone, 2 with buprenorphine, and 69 were in counselling without medication. The EtG-I results were compared with an established GC/MS quantitative method (EtG-MS) in order to assess the diagnostic performance of the EtG rapid test.

## Materials and methods

### Study participants

The study was conducted following the Helsinki Declaration of 1975 as revised in 1983. It was approved by Bioethics Review Board of the University of Perugia (Protocol 2012-006R).

After giving informed consent, 119 subjects (85 males, 34 females) who were registered with the Public Service for Drug Dependence Treatment provided a urine sample for EtG on site rapid determination. Samples were then placed in self-sealing specimen bags and sent by means of a secure custody chain to the forensic laboratory for GC/MS quantitative analysis. Details of each patient's sex, age, duration of self-reported alcohol abuse, drug abuse, and rehabilitation therapy are reported in Table 1. Age distribution: 17 subjects were under 20 years old, 40 were aged 20–30, 27 were between 30–40 years old, 22 between 40–50, and 13 between 50–60. Drug treatments and abuse patterns: 48 patients were in maintenance therapy with methadone and 2 with buprenorphine; 69 received counselling therapy without medication. Of them 10.4% had been methadone clients for under one year, 20.8% for 1 to 3 years, 68.8% for over 3 years. Buprenorphine maintenance lasted less than a year in 1 patient and more than 3 years in another. 55/119 (46.2%) patients were polydrug abusers (opiate, cocaine, amphetamine and benzodiazepines), 28/119 (23.5%) were heroin addicts; 13/119 (11%) were cocaine users, 23/119 (19.3%) were outpatients who had been completely rehabilitated from drug abuse for over 1 year.

### Chemicals, reagents and standards

All chemicals were of analytical grade. EtG and its deuterated analogue ethylglucuronide D5 (EtG-D5) were purchased from Chemical Research 2000 srl

(Rome, Italy). The derivatizing agent N-methyl-N-trimethylsilyltrifluoroacetamide (MSTFA) with 1% TMCS (trimethylchlorosilane) was obtained from Sigma Aldrich (Milan, Italy).

#### EtG immunochemical test

The Nal von Minden Drug-Screen Single Rapid Test (Regensburg, Germany) is an *in vitro* POCT diagnostic device. The rapid test uses the enclosed EtG color card and is equipped with an integrated process control. The control line is the result of an independent antigen/antibody reaction and must always appear independently of drug and metabolite concentrations in the sample. After being applied in the immersion test area, the urine sample moves along the test strip by capillary action, along with free, gold-conjugated antibodies that are located near the immersion area. Development of a red control line indicates the validity of the test. The standard cut-off was set at 500 ng/ml, in accordance with SAMHSA guidelines (SAMHSA, 2011).

#### GC/MS conditions and validation of the method

A GC/MS (Focus DSQ, Thermo Electron Corp., Milano, Italy) operating in electron impact mode (70 eV) with an Equity 5 capillary column, 30 m×0.25 mm×0.25 mm film thickness, was used at the following temperature program: isothermal mode for 2 min at 60 °C, then 10 °C/min to 200 °C, 15 °C/min to 250 °C, 30 °C/min to 280 °C, 5 min isotherm. Column helium flow was 1 ml/min. The injector and transfer lines were maintained at 250 °C and 280 °C, respectively. After protein removal by 100 µl 3 mol/L hydrochloric acid, a 1 µl urine sample was cleaned up through a solid phase extraction column and then derivatized with MSTFA + 1% TMCS to yield the trimethylsylil (TMS) derivative of EtG (EtG-TMS) and EtG-D5 (EtG D5-TMS). 1 µl of this urine extract was injected into the GC/MS in split-less mode. Acquisition in selected ion monitoring (SIM) mode was performed by choosing 3 ions for each compound: EtG-TMS (217-204-147 *m/z)*, EtG D5-TMS (222-209-152 *m/z*). Target ions were underlined and used for quantification.

The method was validated for selectivity, linearity and sensitivity, precision and accuracy, using samples from teetotallers whose histories of total abstinence from ethyl alcohol were available. Seven urine samples were analyzed in order to identify any endogenous interferents. To quantify EtG, three curves were generated at low (50–250 ng/ml), medium (250–500 ng/ml), and high (500–1000 ng/ml) concentrations. The limit of quantification (LOQ) was defined as the lowest concentration with an accuracy with a relative standard deviation (RSD) <20%. The limit of detection (LOD) was defined as signal to noise ratio equal to 5. Accuracy and precision were assessed by analyzing quality control (QC) samples at concentrations of 50 (LOQ), 300, 600 and 1000 ng/ml. Five replicates of each standard were analyzed on five non-consecutive days. Comparing analyses of 5 extracted and 5 non-extracted spiked urine samples at QC concentrations determined the % relative extraction recoveries.

#### Statistical analysis

For all variables that were evaluated in this study Pearson's chi-squared test ($\chi^2$) estimated chance probability of any inter-set differences. For tests on the equality of means, unpaired two-sample Student's *t*-test was used (Gleason, 1999). The diagnostic performance of the EtG-I test was evaluated by Receiver Operating Characteristic (ROC) curve analysis, which defined levels of diagnostic test accuracy, specificity and sensitivity and the optimal cut-off (via the Liu method) (Liu, 2012).

All estimates were conducted using the statistical program STATA 14.2 (Stata Corp ltd, College Station, Texas, USA).

## Results and discussion

Patients who were registered at the Public Health Service for Drug Dependence Treatment provided 119 urine samples which were tested for EtG using a POCT immunochemical test at the 500 ng/ml cut-off level. POCT devices provide rapid, relatively accurate presumptive results, which may be very useful during medical consultation in several medical settings and improve patient management (George & Braithwaite, 2002). EtG-I qualitative results were evaluated against a fully validated GC/MS method, according to international forensic guidelines (Peters *et al.*, 2007).

Linearity of the GC/MS method was determined in the 50–1000 ng/ml range by the least squares regression method, with $R^2$ over 0.99. The LOD and LOQ were 10 and 50 ng/ml, respectively. The intra- and inter- day precision and RSD were always below 10% in QC samples and below 20% for the LOQ. No interferent peak was observed in any sample at the EtG and EtGD5 retention time, demonstrating that the method provided good selectivity. Accuracy, calculated as bias in the difference between expected QC concentration and measured QC concentration, was below 10%. EtG extraction recovery from urine was 80%.

Having thus successfully validated the GC/MS method, quantitative data analysis assessed demographic and alcohol intake features within the study population (Figure 1). Urinary EtG-I was above the 500 ng/ml cut-off in 87/119 (73.1%) patients. A significant relationship emerged between sex and drinking (Figure 1A), with women drinking less alcohol than men (520.6 mean urinary EtG in women vs 712.5 ng/ml in men (***$p<0.001$).

A positive correlation was found between alcohol intake and age. The mean urinary EtG was 422.4 ng/ml in subjects under 20 years old, 626.3 ng/ml in the 20–30 age group, 612.2 in the 30–40 age group, 845.5 in the 40–50 age group, and 783.84 in the 50–60 year-old subjects (Figure 1B). Thus alcohol consumption was highest in the 40–50 year olds (*$p<0.05$) in the study population. Quantitative data analysis by GC/MS showed high urinary EtG levels in subjects in counselling therapy without medication (687 ng/mg). They were not significantly different from levels in patients receiving methadone for under 1 year (520 ng/ml), for 1–3 years (617 ng/ml) and for

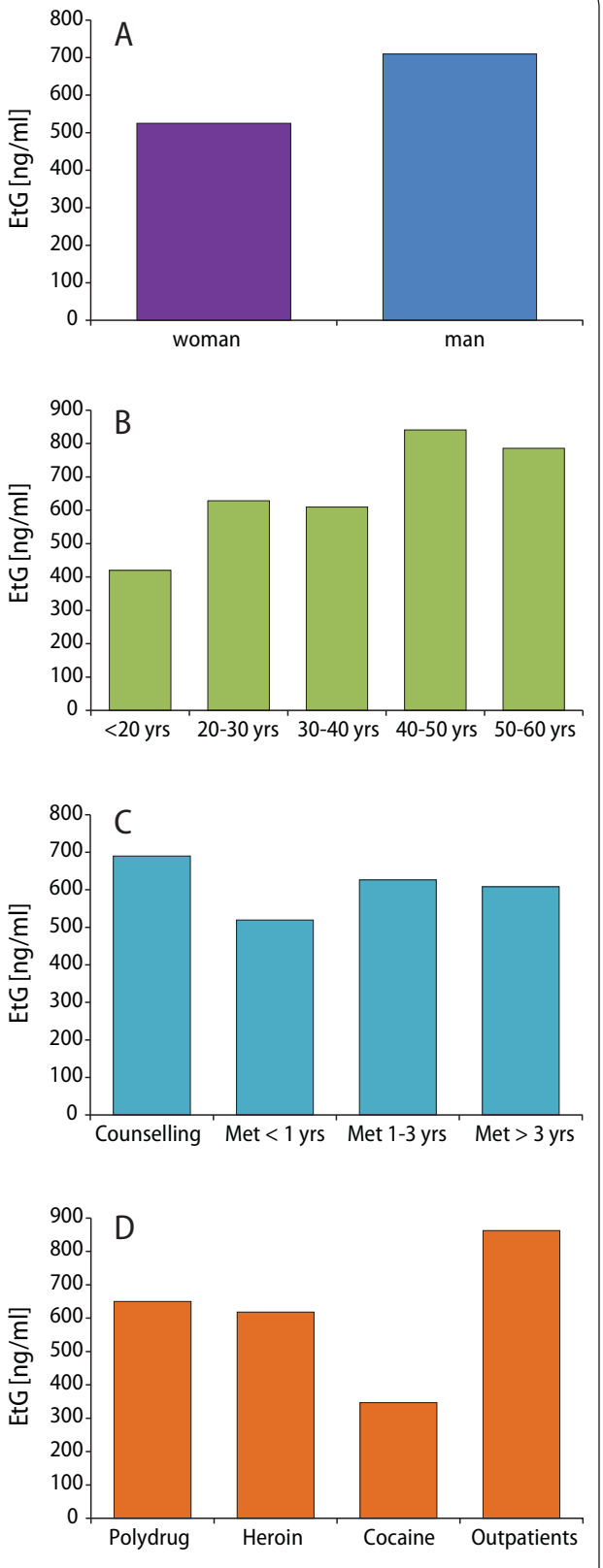


**Figure 1.** Mean EtG urinary values in men and women (**A**), in subjects under 20 years of age and in the 20–30, 30–40 and 40–50 age-groups (**B**), subjects in counselling therapy compared with patients receiving methadone (Met) for periods comprising <1 year , 1–3 years and >3 years (**C**), in polydrug abusers compared with heroin and cocaine users and output patients (**D**).

over 3 years (604.5 ng/ml). Concurring with most studies (Soyka, 2015; Srivastava *et al.*, 2008; Hartzler *et al.*, 2010; Nyamathi *et al.*, 2009), our data indicate that opioid maintenance therapy does not change alcohol consumption (Figure 1C). Although cocaine users are commonly known to consume alcoholic beverages (Pennings *et al.*, 2002), few cocaine abusers (13/119) were included in the present investigation, which may account for their low EtG levels compared with other groups in the study population.

The mean urinary EtG concentration was 643.7 ng/ml in polydrug abusers. It was 618.1 ng/ml in heroin addicts, 343.6 ng/ml in cocaine users and 859.2 ng/ml in outpatients who had been completely rehabilitated from drug abuse for over a year ($p$<0.001) but who still had a strong tendency to use psychoactive substances like tobacco or alcohol (Figure 1D).

Of the urine samples, 32/119 tested negative for EtG-I, 18 were confirmed as true negatives by Etg-MS at the LOQ of 50 ng/ml, 14 were found positive by GC/MS analysis at a value below the EtG-I test cut-off of 500 ng/ml. No false positive results to EtG-I test were identified by EtG-MS.

The present study showed the EtG-I test perfectly identified alcohol consumers with EtG concentrations over the 500 ng/ml cut-off. At the empirical estimated cut-off of 375 ng/ml, the EtG-I test still maintained over 93% specificity, showing it could yield positive results above 375 ng/ml with a high level of specificity. Breaking down these findings by gender and age, the EtG-I test showed greater sensitivity for women and individuals under 30 years old, with the optimal cut-offs emerging as 325 and 310 ng/ml, respectively. Accounting for these low cut-offs is the fact that the lowest urinary EtG concentrations in all the study population categories were found in women and people under 30 years old. Overall, only 14 individuals (12%) in our sample of 119 subjects were not classified correctly as positive (not 0), but with levels below 500 ng/ml which was the standard sensitivity of the test.

The average consumption of the incorrectly classified subjects was 171 ng/ml with a standard deviation of 50, a minimum of 100 and a maximum of 250 ng/ml with a 95% confidence interval, exclusive use of the EtG-I test in question can be estimated to lead to a misclassification error of 6.5% to 18.5%.

Since good levels of agreement emerged between the EtG-I and the EtG-MS test, which was conducted according to international forensic guidelines, EtG-I at the 500 ng cut-off appears to be a reliable objective measure of alcohol intake suitable for monitoring moderate-to-high alcohol use in non-forensic settings. Heavy drinking, in fact, warrants careful monitoring, since it has been associated with quality of life impairments (Nyamathi *et al.*, 2009).

EtG appears a reliable and relatively long-term marker of ethanol exposure. Measurement of this minor ethanol metabolite has yet to realize its full potential as a valuable asset in treatment programs involved in managing alcohol e/o drug abuse disorders (Jatlow & O'Malleys, 2010). The high sensitivity of EtG for recent drinking is evident from

**Table 1.** Study participants.

| Patient | Age range | Sex | Years of alcohol abuse (Self report) | Abused Drugs | Duration of therapy with methadone (years) | Duration of therapy with buprenorphine (years) | EtG-I | ETG-GC-MS (ng/ml) |
|---|---|---|---|---|---|---|---|---|
| 1 | 20–30 | M | 8 | P | <1 | N.S. | POS | 600 |
| 2 | 20–30 | M | 10 | P | 1–3 | N.S. | POS | 800 |
| 3 | 40–50 | M | 30 | H | N.S | > 3 | POS | 1300 |
| 4 | 20–30 | F | 8 | P | 1–3 | N.S. | POS | 500 |
| 5 | 30–40 | F | 20 | H | > 3 | N.S. | NEG | 100 |
| 6 | 40–50 | M | 2 | P | >3 | N.S. | POS | 850 |
| 7 | 40–50 | M | 2 | P | >3 | N.S. | POS | 700 |
| 8 | 30–40 | M | 17 | P | 1–3 | N.S. | NEG | 150 |
| 9 | 40–50 | M | 20 | H | > 3 | N.S. | NEG | 220 |
| 10 | 20–30 | F | 2 | H | >1 | N.S. | NEG | NEG |
| 11 | 30–40 | M | 2 | P | >3 | N.S. | POS | 800 |
| 12 | 20–30 | F | 8 | P | >1 | N.S. | NEG | NEG |
| 13 | >20 | F | 4 | H | 1–3 | N.S. | POS | 700 |
| 14 | 20–30 | M | 19 | P | 1–3 | N.S. | POS | 1000 |
| 15 | 20–30 | M | 10 | P | >3 | N.S. | NEG | 200 |
| 16 | 50–60 | M | 35 | P | 1–3 | N.S. | POS | 1300 |
| 17 | 30–40 | M | 20 | P | N.S. | <1 | POS | 750 |
| 18 | 20–30 | F | 10 | P | >3 | N.S. | POS | 800 |
| 19 | <20 | F | 2 | P | 1–3 | N.S. | NEG | NEG |
| 20 | 20–30 | F | 10 | P | >3 | N.S. | POS | 820 |
| 21 | <20 | F | 4 | P | N.S. | N.S. | POS | 550 |
| 22 | 30–40 | M | 21 | H | >3 | N.S. | POS | 1200 |
| 23 | 40–50 | M | 20 | H | >3 | N.S. | POS | 980 |
| 24 | 40–50 | M | 30 | P | > 3 | N.S. | POS | 750 |
| 25 | 50–60 | M | 5 | P | >3 | N.S. | POS | 600 |
| 26 | 30–40 | F | 20 | H | > 3 | N.S. | NEG | NEG |
| 27 | 40–50 | M | 20 | P | > 3 | N.S. | NEG | NEG |
| 28 | 50–60 | M | 15 | H | 1–3 | N.S. | POS | 820 |
| 29 | 40–50 | M | 20 | P | <1 | N.S. | POS | 1200 |
| 30 | 40–50 | F | 20 | H | >3 | N.S. | POS | 950 |
| 31 | 30–40 | M | 10 | H | >3 | N.S. | POS | 700 |
| 32 | 40–50 | F | 20 | H | > 3 | N.S. | POS | 850 |
| 33 | 40–50 | F | 18 | H | > 3 | N.S. | POS | 1100 |
| 34 | 40–50 | F | 28 | P | N.S. | N.S. | POS | 750 |
| 35 | 20–30 | F | 15 | C | N.S. | N.S.. | POS | 680 |
| 36 | 30–40 | M | 20 | H | > 3 | N.S. | POS | 500 |
| 37 | <20 | M | 2 | P | N.S. | N.S. | POS | 600 |
| 38 | 50–60 | M | 30 | H | > 3 | N.S. | NEG | 250 |
| 39 | 20–30 | F | 5 | P | N.S. | N.S. | POS | 600 |
| 40 | 20–30 | M | 14 | C | N.S. | N.S. | POS | 700 |
| 41 | 20–30 | F | 10 | H | > 3 | N.S. | POS | 500 |
| 42 | 50–60 | F | 30 | C | N.S. | N.S. | NEG | NEG |
| 43 | 30–40 | M | 20 | C | N.S. | N.S. | POS | 500 |
| 44 | 40–50 | M | 23 | C | N.S. | N.S. | NEG | NEG |
| 45 | 20–30 | F | 10 | H | 1–3 | N.S. | POS | 900 |
| 46 | 40–50 | F | 27 | H | > 3 | N.S. | POS | 950 |
| 47 | 30–40 | M | 20 | H | > 3 | N.S. | NEG | NEG |
| 48 | 20–30 | M | 14 | C | N.S. | N.S. | NEG | NEG |
| 49 | 50–60 | M | 34 | H | > 3 | N.S. | POS | 700 |
| 50 | 40–50 | M | 26 | P | N.S. | N.S. | POS | 1100 |
| 51 | 20–30 | F | 5 | H | > 3 | N.S. | POS | 850 |
| 52 | <20 | M | 3 | P | N.S. | N.S. | POS | 600 |
| 53 | <20 | M | 2 | P | N.S. | N.S. | NEG | NEG |

Legend: P: polydrug abusers; H: heroin addicts; C: cocaine users, N: currently no drug use; N.S.: not in use; EtG-I: ethylglucoronide immunochemical test; POS: positive; NEG: negative

**Table 1.** *Continued*

| Patient | Age range | Sex | Years of alcohol abuse (Self report) | Abused Drugs | Duration of therapy with methadone (years) | Duration of therapy with buprenorphine (years) | EtG-I | ETG-GC-MS (ng/ml) |
|---|---|---|---|---|---|---|---|---|
| 54 | 30–40 | M | 20 | H | > 3 | N.S. | POS | 1150 |
| 55 | 20–30 | M | 9 | C | N.S. | N.S. | POS | 600 |
| 56 | 20–30 | F | 8 | H | > 3 | N.S. | POS | 700 |
| 57 | 20–30 | M | 10 | P | N.S. | N.S. | POS | 980 |
| 58 | 20–30 | F | 15 | C | N.S. | N.S. | POS | 800 |
| 59 | <20 | M | 2 | P | > 1 | N.S. | POS | 800 |
| 60 | 20–30 | M | 10 | H | > 3 | N.S. | POS | 730 |
| 61 | <20 | F | 2 | N | N.S. | N.S. | NEG | NEG |
| 62 | <20 | M | 32 | C | N.S. | N.S. | NEG | NEG |
| 63 | <20 | F | 1 | P | N.S. | N.S. | NEG | NEG |
| 64 | 30–40 | M | 20 | H | > 3 | N.S. | NEG | 250 |
| 65 | 30–40 | F | 18 | H | > 3 | N.S. | NEG | NEG |
| 66 | 20–30 | F | 6 | H | N.S. | N.S. | POS | 740 |
| 67 | <20 | M | 10 | C | N.S. | N.S. | NEG | 180 |
| 68 | <20 | F | 3 | P | N.S. | N.S. | NEG | NEG |
| 69 | 40–50 | F | 25 | P | > 3 | N.S. | NEG | NEG |
| 70 | 20–30 | M | 10 | P | 1–3 | N.S. | NEG | NEG |
| 71 | 20–30 | M | 17 | C | N.S. | N.S. | POS | 1000 |
| 72 | 50–60 | M | 32 | H | > 3 | N.S. | POS | 900 |
| 73 | 20–30 | M | 5 | C | N.S. | N.S. | NEG | NEG |
| 74 | 40–50 | M | 24 | P | > 3 | N.S. | POS | 850 |
| 75 | 20–30 | M | 2 | N | N.S. | N.S. | POS | 850 |
| 76 | 50–60 | M | 25 | N | N.S. | N.S. | POS | 1250 |
| 77 | 20–30 | M | 4 | P | N.S. | N.S. | POS | 750 |
| 78 | 30–40 | M | 7 | N | N.S. | N.S. | POS | 1200 |
| 79 | >20 | M | 2 | N | N.S. | N.S. | POS | 650 |
| 80 | 20–30 | M | 10 | P | N.S. | N.S. | POS | 650 |
| 81 | 20–30 | M | 10 | P | N.S | N.S. | POS | 1300 |
| 82 | 20–30 | F | 5 | P | N.S. | N.S. | POS | 920 |
| 83 | 20–30 | M | 8 | N | N.S. | N.S. | POS | 800 |
| 84 | 30–40 | M | 5 | N | N.S. | N.S. | NEG | 200 |
| 85 | 20–30 | M | 5 | P | N.S. | N.S. | NEG | 150 |
| 86 | 30–40 | M | 10 | P | N.S. | N.S. | POS | 600 |
| 87 | 50–60 | M | 20 | N | N.S. | N.S. | POS | 800 |
| 88 | 40–50 | M | 15 | N | N.S | N.S. | POS | 1200 |
| 89 | <20 | M | 3 | N | N.S. | N.S. | POS | 800 |
| 90 | 40–50 | M | 20 | N | N.S. | N.S. | POS | 1000 |
| 91 | <20 | M | 5 | P | N.S. | N.S. | POS | 900 |
| 92 | 30–40 | M | 10 | N | N.S. | N.S. | POS | 1000 |
| 93 | <20 | M | 2 | P | N.S. | N.S. | POS | 700 |
| 94 | 30–40 | M | 9 | P | N.S. | N.S. | POS | 600 |
| 95 | 20–30 | M | 10 | P | N.S. | N.S. | POS | 1400 |
| 96 | 30–40 | F | 6 | P | N.S. | N.S. | POS | 920 |
| 97 | 30–40 | M | 7 | N | N.S. | N.S. | POS | 900 |
| 98 | 30–40 | M | 5 | N | N.S. | N.S. | NEG | 200 |
| 99 | 20–30 | M | 5 | P | N.S. | N.S. | NEG | 150 |
| 100 | 30–40 | M | 10 | P | N.S. | N.S. | POS | 940 |
| 101 | 50–60 | M | 20 | N | N.S. | N.S. | POS | 820 |
| 102 | 40–50 | M | 15 | N | N.S. | N.S. | POS | 1250 |
| 103 | 20–30 | F | 5 | P | N.S. | N.S. | POS | 800 |
| 104 | 20–30 | M | 6 | N | N.S. | N.S. | POS | 1000 |
| 105 | 30–40 | M | 5 | N | N.S. | N.S. | NEG | 100 |
| 106 | 20–30 | M | 5 | P | N.S. | N.S. | NEG | 130 |

Legend: P: polydrug abusers; H: heroin addicts; C: cocaine users, N: currently no drug use; N.S.: not in use; EtG-I: ethylglucoronide immunochemical test; POS: positive; NEG: negative

**Table 1.** *Continued*

| Patient | Age range | Sex | Years of alcohol abuse (Self report) | Abused Drugs | Duration of therapy with methadone (years) | Duration of therapy with buprenorphine (years) | EtG-I | ETG-GC-MS (ng/ml) |
|---|---|---|---|---|---|---|---|---|
| 107 | 30–40 | M | 10 | P | N.S. | N.S. | POS | 900 |
| 108 | 50–60 | M | 18 | N | N.S. | N.S. | POS | 800 |
| 109 | 40–50 | M | 10 | P | N.S. | N.S. | POS | 1400 |
| 110 | 20–30 | F | 5 | P | N.S. | N.S. | NEG | 120 |
| 111 | 30–40 | M | 9 | P | N.S. | N.S. | POS | 940 |
| 112 | 50–60 | M | 22 | C | N.S. | N.S. | POS | 1000 |
| 113 | 50–60 | M | 15 | N | N.S. | N.S. | POS | 950 |
| 114 | 20–30 | M | 2 | P | N.S. | N.S. | POS | 830 |
| 115 | 40–50 | M | 24 | N | N.S. | N.S. | POS | 1200 |
| 116 | 20–30 | M | 4 | P | N.S. | N.S. | POS | 700 |
| 117 | 30–40 | F | 6 | N | N.S. | N.S. | POS | 1100 |
| 118 | <20 | M | 2 | N | N.S. | N.S. | POS | 700 |
| 119 | 30–40 | M | 10 | P | N.S. | N.S. | POS | 830 |

Legend: P: polydrug abusers; H: heroin addicts; C: cocaine users, N: currently no drug use; N.S.: not in use; EtG-I: ethylglucoronide immunochemical test; POS: positive; NEG: negative

the observation that even a very low dose (~7 g) of ethanol is detected in the urine after 6 h and for up to 4 days after the last intake (Stephanson *et al.*, 2002).

## Conclusion

POCT EtG-I urine measurement can be considered a routine test for monitoring recent alcohol intake. Rates of agreement between EtG-I and EtG-MS results confirm that EtG-I objectively detects recent moderate-to-high alcohol intake. It may prove a helpful tool in interventions, such as drug rehabilitation programs, that aim at reducing hazardous/harmful alcohol use, particularly when alcohol plays a major role in the overall pattern of multiple substance use. Such programs might be comfortable with EtG 500ng/ml cut offs, especially if testing is frequent, in a manner analogous to urinary drug abuse screening. EtG-I is fast, inexpensive and technically suitable for point of care or on-site testing in all non-forensic settings, with the purposes of diagnosis, treatment, and the promotion of long-term recovery from excessive alcohol consumption.

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### *Conflict of interest statement*

*We declare that there is no conflict of interest.*

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