Abstinence monitoring of suspected drinking drivers: ethyl glucuronide in hair versus CDT


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Abstract

Objective: Ethyl glucuronide (EtG) determinations in the hair of self-reported teetotallers were reviewed and compared with CDT blood tests (by immunochemistry and HPLC). Methods: A retrospective study was carried out on 154 people whose fitness to drive had to be assessed because of the suspicion of relevant alcohol problems. Results: EtG was detected in 55% of the hair samples, and abstinence thus disproved. In two thirds (67%) of these cases, alcohol consumption was even shown to be excessive (EtG values >30 pg/mg). Of the EtG-positive subjects 54% and 82% had CDT values within the reference range by immunochemistry and HPLC, respectively. 39% of the EtG-negative subjects had increased immunochemical CDT values; in contrast, 96% had HPLC CDT values within the normal range. Conclusions: EtG analysis in hair is a useful tool for assessing fitness to drive in suspected drinking drivers; compared to CDT values it provides a direct and unequivocal marker for reliable abstinence monitoring over a period of several months, depending on the length of the hair.
Abstinence monitoring of suspected drinking drivers: ethyl glucuronide in hair versus CDT

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ABSTRACT

Objective: Ethyl glucuronide (EtG) determinations in the hair of self-reported teetotallers were reviewed and compared with CDT blood tests (by immunochemistry and HPLC).

Methods: A retrospective study was carried out on 154 people whose fitness to drive had to be assessed because of the suspicion of relevant alcohol problems.

Results: EtG was detected in 55% of the hair samples, and abstinence thus disproved. In two thirds (67%) of these cases, alcohol consumption was even shown to be excessive (EtG values >30 pg/mg). Of the EtG-positive subjects 54% and 82% had CDT values within the reference range by immunochemistry and HPLC, respectively. 39% of the EtG-negative subjects had increased immunochemical CDT values; in contrast, 96% had HPLC CDT values within the normal range.

Conclusions: EtG analysis in hair is a useful tool for assessing fitness to drive in suspected drinking drivers; compared to CDT values it provides a direct and unequivocal marker for reliable abstinence monitoring over a period of several months, depending on the length of the hair.

Keywords

Ethyl Glucuronide; Hair Analysis; Fitness to Drive; Alcohol Abuse; Abstinence Monitoring
INTRODUCTION

One of the most frequent tasks in traffic medicine today is the expert appraisal of road users who have drinking problems affecting their fitness to drive. Experts face the challenge of proving suspected chronic excessive alcohol consumption as unequivocally as possible, or reliably monitoring claimed or enforced abstinence. In addition to a full review of the records, in-depth questioning and physical examination, the work-up in such cases includes carrying out relevant laboratory blood tests: GGT, AST, ALT, MCV and CDT (carbohydrate-deficient transferrin). One thing that all these indirect, more or less sensitive and specific, alcohol markers have in common is that they cover only a limited period of time and may be affected by other factors, such as severe hepatobiliary disease, metabolic disorders, and genetic variants. Whenever elevated values are found in the assessment of fitness to drive, despite claims of long-term abstinence from alcohol, discussions inevitably arise between the person concerned, that person’s own doctor, the legal representative, and the expert. It would therefore be useful to have a direct alcohol marker with informative value over a longer retrospective period.

0.02% to 0.06% of the ethyl alcohol consumed is metabolised to ethyl glucuronide (EtG) by non-oxidative conjugation with co-factor UDP glucuronic acid in the liver (Dahl et al., 2002). Ethyl glucuronide is a stable water-soluble substance that is produced only after drinking ethyl alcohol and has already been found in several body fluids and tissues. In 1995, EtG was demonstrated in hair after the regular consumption of small quantities of alcohol (Schloegl et al., 2006; Skopp et al., 1995; Skopp et al., 2000; Wurst and Metzger, 2002; Yegles et al., 2004). Determination of EtG in hair offers a comparatively long period of detection. On the basis of scalp hair growing at an average rate of 1 cm/month, repeated alcohol consumption can be confirmed over the preceding months, depending on the length of the hair. On the other hand, because it will not show a solitary episode of modest alcohol consumption, EtG hair analysis is not a suitable method for screening. Various methods may be used for the analysis, such as liquid chromatography/mass spectrometry and gas chromatography/mass spectrometry (Pragst and Yegles, 2007). Some people who reported drinking alcohol regularly showed no evidence of EtG in the hair (Wurst et al., 2003); that is to say, negative EtG findings are not proof of abstinence. On the other hand, a positive result can be taken as confirmation of alcohol
consumption within the period of detection (Yegles et al., 2004). A statistically significant correlation has been found between the EtG concentration in the hair segment and the amount of alcohol consumed (Appenzeller et al., 2007).

Since the middle of 2005, the Institute of Legal Medicine in Zurich has added the determination of ethyl glucuronide (EtG) in hair to the standard routine analyses in cases of suspected alcohol problems. The aim of our study was to compare ethyl glucuronide (EtG) results in hair of self-reported teetotallers with CDT blood tests (by immunochemistry and HPLC).

**METHODS**

A retrospective study was carried out on 154 people whose fitness to drive had to be assessed because of the suspicion of relevant alcohol problems. EtG determinations in hair, together with CDT measurements by an immunochemical method (immunoturbidimetry) and by HPLC, were carried out in all subjects as part of the traffic medicine examination. CDT blood tests carried out in other laboratories during the period of hair growth were also taken into consideration in the overall analysis. The CDT cut-off was taken as <2.6% for the immunochemical method used and as <1.77% for HPLC. All subjects claimed that they had not drunk any alcohol at all during the study period, which ranged from a few weeks to several months, depending on the length of the hair. The study population included 132 men (86%) and 22 women (14%).

In 143 of the subjects, one or two small tufts of underlying scalp hair were tied with thread and cut off close to the skin with a pair of scissors. In 11 cases, secondary body hair (from the chest or calves) was shaved off with a disposable razor. The samples were wrapped in aluminium foil, put into resealable plastic bags, and sent to the National Public Health Laboratory, University of Luxemburg, for analysis. GC/MS-NCI was carried out to determine EtG in the hair, using the method first described by Yegles et al. (2001) and modified by Kerekes et al. (2009).

As the EtG concentration in hair usually correlates with the amount of alcohol consumed (Appenzeller et al., 2007), it is particularly important to establish a cut-off level (the border between social drinking and alcohol abuse, i.e. regular and excessive consumption of alcohol). The toxicology laboratory in Luxembourg used the following cut-offs:
• EtG concentration in hair <7 pg/mg (= negative result): No regular consumption of alcohol, abstinence cannot be disproved, occasional alcohol consumption is possible

• EtG concentration in hair 7 - 30 pg/mg: Social drinking (20 to 40 g alcohol/day); abstinence is clearly disproved

• EtG concentration in hair >30 pg/mg: Excessive and regular alcohol consumption/abuse; abstinence is clearly disproved

RESULTS

The study population (n=154) was divided into subjects with positive results (>7 pg/mg hair (n=84) and those with negative EtG findings (<7 pg/mg hair) (n=70). In both groups EtG findings were compared with the results of CDT testing (by immunochemistry and HPLC). If several CDT measurements were available, the overall classification of CDT in the normal range or elevated CDT was made according to the number of results that fell into each category. In borderline cases, we took the CDT value determined as part of the traffic medicine examination. Only the concentrations obtained from scalp hair were included in the quantitative EtG analysis.

Positive EtG findings compared with Immunochemical/HPLC CDT values

EtG findings in the hair samples were positive in 84 out of the 154 self-reported teetotallers, i.e. abstinence from alcohol was disproved in a good half of the study population. However, only 39 of these 84 subjects had a CDT value above the upper limit of normal using the immunochemical method. Moreover, when measured by HPLC, only 15 subjects had a CDT value above the cut-off (figure 1).

Negative EtG findings versus Immunochemical/HPLC CDT values

Hair analysis for EtG was negative in 70 of the 154 self-reported teetotallers, i.e. the findings were compatible with abstinence in 45% of the cases. However, 27 subjects had an immunochemical CDT value above the cut-off although an elevated HPLC CDT value was recorded in only 3 cases (figure 2).
Quantitative EtG results

The quantitative EtG results were divided into five concentration ranges as shown in figure 3. 26 subjects (33%) had EtG concentrations of less than 30 pg/mg; this range has been classed as social drinking. The EtG concentration was more than 30 pg/mg in 67% of subjects for which a regular excessive alcohol consumption has been assumed: 30-100 pg/mg in 41 subjects (52%); 100-200 pg/mg in 8 subjects (10%); between 200 and 300 pg/mg in two subjects (2.5%); and over 300 pg/mg in two further subjects (2.5%).

DISCUSSION

In assessing fitness to drive in cases where there is a question of alcohol problems, laboratory testing – usually determination of the relevant alcohol-related parameters CDT, GGT, AST, ALT and MCV in the blood – provides an important piece of the puzzle in the assessment of alcohol consumption or in proving/disproving self-reported abstinence. If someone is declared unfit to drive because of alcohol problems (abuse, dependence), the traffic medicine guidelines in Switzerland require long-term complete abstinence from alcohol (for at least 6 months, and possibly for 12 months). To check that the person has not been drinking, tests for the indirect alcohol markers are done about every 6-8 weeks; the blood samples are usually taken by the general practitioner (Seeger, 2005). CDT has been shown to be specific and sensitive, but results are only above the upper limit of normal after the consumption of more than 60 g alcohol daily for more than two weeks, so it often does not detect short periods of high intake (Arndt, 2001). In addition, the CDT concentration does not rise in a certain group of people who drink alcohol regularly (false negative result in non-responders) (Barbon-Jermini et al., 1999). The use of different methods for CDT assays (immunochemistry/ HPLC) also gives rise to discussion because of different sensitivities, specificities, and references ranges. In 2005, to improve this unsatisfactory situation, the Institute of Legal Medicine, University of Zurich, introduced hair analysis for EtG into the assessment of fitness to drive. In contrast to the other routine parameters, EtG is a direct alcohol marker and requires a non-invasive procedure to obtain the sample; scalp hair should be taken whenever possible. Factors such as severe liver disease or medications seem not to give false positive results; the matrix also provides information over a longer period of time (depending on the length of the hair). In addition, information about the history of
alcohol consumption may be gained by looking at different segments of the sample.

In this investigation, 154 subjects claimed that they had been teetotal during the study period or had drunk very little alcohol (not more than two beers a month). Theoretically, negative EtG findings were to be expected in all of these cases. Surprisingly though, about half of them (n=84) had EtG in the sample tested – a finding that clearly and strongly refuted the claim of abstinence. As isolated episodes of drinking are not detected by analysing hair for EtG, it has to be assumed that these subjects had been repeatedly consuming not negligible quantities of alcohol during this time. The distribution of the EtG concentrations ranges found in the scalp hair (n=79) was also somewhat astonishing: 52% had EtG concentrations between 30 pg/mg and 100 pg/mg, i.e. members of this group claiming abstinence must have been drinking alcohol not only regularly but also in excessive amounts. EtG levels were even higher than 100 pg/mg in 15% of these self-reported teetotallers.

When comparing the EtG findings in the hair with the CDT concentrations in the blood, it has to be remembered that hair analysis does not cover a period of about 14 days prior to sampling, while CDT values generally reflect the alcohol consumption of the past two weeks. The finding that more than half the subjects with positive EtG findings (45/84) had an unremarkable immunochemical CDT result may be explained by the fact that, knowing they had to undergo a medical examination, they reduced their alcohol intake for some time beforehand. HPLC CDT determination was within the normal range in as many as 82% of cases (69/84); this could also mean that the upper limit for CDT by HPLC has been set too high. Not taking the different periods of detection into account, if the check on abstinence had been carried out on the basis of CDT determination alone, 54% and 82% of the subjects would have shown no evidence of excessive alcohol consumption by immunochemistry and HPLC, respectively. In other words, the abstinence claimed would not have been disproved.

A good third of the subjects with negative EtG findings (27/70) had an elevated immunochemical CDT value. On looking at this group more closely, it turned out that five subjects had liver disease (end-stage hepatic failure, alcohol-induced liver disease, history of hepatitis B) and for this reason had falsely elevated CDT levels despite being teetotal; the EtG hair analysis could confirm the abstinence they claimed. Four subjects had cosmetically treated hair (bleached, toned, dyed, double process), which may lead to a reduction of as much as 75% in the EtG concentration or to an EtG level below the detection limit (Yegles et al., 2004); in these cases, exogenous influences caused the semblance
of abstinence. For further four subjects only secondary hair (from the chest or legs) was taken for testing; EtG findings in secondary hair generally have to be interpreted with caution (Kerekes et al., 2009). Correlating with the negative EtG findings, HPLC CDT values were in the normal range in 96% of the cases – and it should be remembered that CDT levels measured by HPLC show the greatest diagnostic specificity. In contrast, three subjects (4%) had elevated CDT levels measured by HPLC as well as by immunochemistry, although once again only secondary hair was available for EtG analysis in two of the cases. Despite the negative hair samples, excessive alcohol consumption and non-compliance with abstinence has to be assumed in these subjects.

Despite certain limitations (no evidence of isolated drinking episodes, about a two-week gap in the period of detection) and stumbling blocks in the interpretation (false negative results in cosmetically treated hair, dilution effect of periodic drinking in long hair samples) determination of EtG in hair is a considerable advance in assessing fitness to drive. Positive EtG findings confirm alcohol consumption directly and unequivocally, which means that alleged or enforced abstinence can be reliably monitored. Depending on the length of the hair long-term drinking patterns over several months previously can be evaluated. In individual cases, determination of EtG in the hair may also contribute to exonerating the subject (negative EtG findings with false positive indirect alcohol markers). Thus, EtG hair analysis should be added to the routine assessment of fitness to drive in cases of alcohol problems, to provide experts with greater certainty in their appraisals.

REFERENCES


Figure 1: Cases with positive EtG findings in hair (>7 pg/mg) and their CDT values by immunochemical and HPLC methods

Figure 2: Cases with negative EtG findings in hair (<7 pg/mg) and their CDT values by immunochemical and HPLC methods

Figure 3: EtG concentrations ranges in scalp hair of 79 self-reported teetotalers