

FAEE Hair Alcohol Test Facts and Experiences

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The detection of illegal drugs has been an established procedure in forensic toxicology since the mid-1990s. Once ingested, opiates, cocaine, cannabinoids and amphetamines are metabolised in the body and enter into the hair root from where they are deposited permanently in the hair in characteristic relationships parent substance/metabolite. As hair grows from the root (at approx. 1 cm per month), these deposits are delayed to a certain extent. The distance from the scalp of the hair portion containing drugs allows us to estimate the approximate time of drug consumption. The deposited substances are relatively stable in the face of external influences such as hair treatments, cosmetics and sunlight. Due to the way in which drugs are deposited in the hair, hair analysis provides us with a detailed view of drug consumption over the past weeks or months. Consumption on a day-to-day basis can be detected by analysing blood and urine samples.

Alcohol is not as easily detectable in hair as other drugs. Ethanol is present in all hair including those of teetotallers, however these traces are products of the environment. Simply being in a pub or laboratory is enough for traces of ethanol to find their way into the hair. So these traces of ethanol do not correlate to alcohol which has been consumed.

In contrast to other drugs consumed, alcohol is not deposited directly in the hair. For this reason the investigation procedure looks for direct products of ethanol metabolism. The main part of alcohol is oxidized in the human body. This means it is released as water and carbon dioxide. One part of the alcohol reacts with fatty acids to produce esters. **The sum** of the concentrations of four of these **fatty acid ethyl esters (FAEEs: ethyl myristate, ethyl palmitate, ethyl oleate and ethyl stearate)** are used as **indicators of the alcohol consumption**. The amounts found in hair are measured in nanograms (one nanogram equals only one billionth of a gram), however with the benefit of modern technology, it is possible to detect such small amounts.

In the mass spectrometer the substances are fired with an electronic beam. Every molecule decomposes into specific fragments. It is possible to determine which substance is present on basis of its molecular weight.

However there is one major difference between most drugs and alcohol metabolites (FAEE) in the way in which they enter into the hair: on the one hand like other drugs FAEEs enter into the hair **via the keratinocytes**, the cells responsible for hair growth. These cells form the hair in the root and then grow through the skin surface taking any substances with them. On the other hand the sebaceous glands produce FAEEs in the scalp and these migrate together with the **sebum** along the hair shaft (Auwärter *et al.*, 2001, Pragst *et al.*, 2004). So these glands lubricate not only the part of the hair that is just growing at 0.3 millimeters per day on the skin surface, but also the more mature

hair growth, providing it with a protective layer of fat.

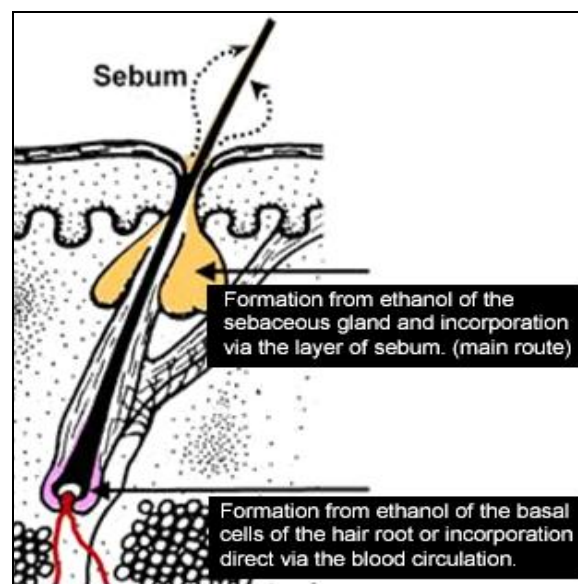


Fig 1: Possible ways of incorporation of FAEEs into hair (based on Auwärter, 2006).

This means the glands supply the whole length of hair with sebum, which in turn carries FAEEs into the hair, resulting in an accumulative increase of FAEEs from proximal to distal hair sections, see next figure (Auwärter *et al.*, 2001).

This phenomenon has one important consequence: whilst most other drugs are mainly stored in the hair via the root alone, allowing toxicologists to establish time-resolved patterns of consumption according to the length of hair provided, this is not possible with alcohol (FAEEs) with respect to previous drinking and abstinence. This would only be possible using alcohol markers which enter the hair solely through the hair root, but these have not yet been discovered.

However there exists a significant correlation between the intensity of the alcohol intake and the concentrations of the FAEEs in hair. Results

between 0.05 and 30 ng/mg were found in hair (Pragst *et al.*, 2006).

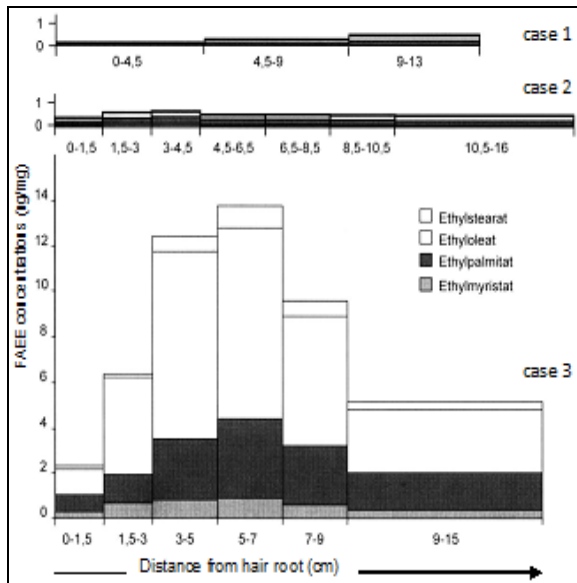


Fig. 2: Profile of FAEE concentrations (based on Pragst, 2004). Case 1 is a strictly abstinent person. Case 2 is a social drinker. Case 3 is a patient in alcohol rehabilitation (self-reporting 60 grams ethanol per day for at least 6 months). The increase in FAEE concentrations from proximal to distal sections up to a length of 5-10 cm found in most cases could be explained by the sebum deposition route. The hair is continuously bathed by sebum, and this leads to an accumulation of the concentrations with increasing age, i.e., distance of hair from skin.

As a consequence of the increasing FAEE concentrations from proximal to distal it is preferable to always analyse, if possible, a standard length of hair. For example analysis of a 1cm hair sample taken close to the scalp will give a much lower result than the analysis of a 6 cm long sample also taken close to the scalp from the same person. For this reason we use as standard - as in all recent studies (since 2001)- **the proximal 0-6 cm segment of head hair in exactly** the same procedure. For samples analysed under these standard conditions the cutoff level is set at 1.0 ng/mg. The same holds true for **body hair of any length** (length is genetically determined, body hair grows more slowly and has different growth cycles, etc.), however not for shorter head hair (e.g.: for only 3 cm we reduce the cut off level to 0.5 ng/mg). The closer the conditions (i.e. hair length) are to our standard procedure, the more certain we can be of the accuracy of the result.

A 6 cm long hair sample can be used to look back 6 months (this is commonly used in Germany to test drivers). If head hair is not available or too short (minimum: 2 cm) underarm, chest, leg and pubic hair may be analysed. Body hair gives us a picture going back up to twelve months.

Frequently Asked Questions

In layman's terms, how much alcohol constitutes alcohol abuse?

Currently, the World Health Organization (WHO) has defined alcohol abuse as the consumption of 60 grams or more of alcohol daily. In the UK, alcohol is based on units to allow drinkers to establish their alcohol consumption. One unit is 8 grams (10 millilitres) of pure alcohol. However, the amount of drink that equates to 60 grams depends on how strong the drink is. On average, one pub measure of wine, one half pint of average strength beer, and one "short" of spirit are all one unit. So seven of these units provide $7 \times 8 = 56$ grams of alcohol. A 750ml bottle of wine with 13% alcohol by volume would contain 9.75 units.

Can we tell if an excessive drinker really has stopped drinking?

After 1-2 months if an excessive alcohol drinker has ceased drinking (i.e. if he is abstinent). In some cases the FAEE values sink after only one month of abstinence to below the cut off level, however at most after two months. Following 3 months of abstinence the values will increasingly liken to those of social drinkers and teetotalers. Please note: For this test we also require, where possible, 6 cm of head hair (standard conditions).

Is hair testing for alcohol an imprecise science?

I would not say that hair testing is an imprecise science. It is subjected to the same rigours and decisions that apply to other branches of testing such as blood alcohol in drivers, for example.

In many areas of medicine and science most testing is done to establish a level and then that level is compared with the best available 'threshold' at the time. To use the above example, drink driving evaluations are carried out against an accepted 'cut off level' which in this case is 35 micrograms of alcohol in breath, 80 milligrams per 100 ml in blood or 108 milligrams in urine, even though those figures do not correlate to each other exactly in any one individual at any one time. In future the cut off level may reduce to 50 milligrams per 100ml of blood. Each country has their own standard, in Scandinavia for example, the driving limit is only 20 mg/100 ml of blood.

Similarly the best thinking on alcohol misuse is currently 60 grams per day (taken every day, or equivalent in binge drinking) but this may be revised by the authorities in future.

Drink driving limits have been established by physical tests which identify response times and concentration levels in individuals compared to alcohol level in their blood.. In practice there is a large amount of 'results scatter' because alcohol

affects different people in different ways, but having a better response to alcohol than one's fellow road users is not a valid defence.

Similarly, the cut off level of 60 grams of alcohol per day has been related to the quantities of hair alcohol markers detected in a very large number of trial subjects. The decision as to what constitutes an alcoholic is not ours, and some people who become classified as excessive consumption by our test will no doubt consider they are not drinking to excess, in the same way that many drivers caught with an excess of alcohol in their blood will argue it improves their driving.

Is there a risk of false positives?

Mitigating circumstances could include a serious abnormality in the metabolism of the donor or abnormal hair growth conditions such as hirsutism, or indeed recent donor hair transplanting. While we would offer constructive advice on such occasions, they are not encountered or required very frequently. Trimega has led the field in this work and is the only organisation worldwide to offer this service coupled with a thorough and considered medical review, and as such is committed to offering as much information and advice as possible to clients.

Can hair treatments affect results?

We have not found any influence from cosmetics and shampoo but there may be occasions when this should be considered (example: hair modeling where the hair is constantly being styled). Recent alcohol application to the scalp for head lice will also not affect the results. Our experience in this field would allow us to offer constructive advice on such occasions.

Why do we use FAEEs and the FAEEs combined with EtG but not EtG alone to indicate alcohol abuse over longer time periods?

EtG alone is still not practicable.

FAEEs (nanogram = one billionth of a gram) appear in hair in almost one order of magnitude higher than (the relevant order of magnitude of) EtG (picogram = one trillionth of a gram). It has been technically possible to measure FAEEs since 1993, whereas the technique for measuring (the relevant range of) EtG is still in its infancy.

In practice, most hair which is sent for analysis has been cosmetically treated in some way (bleached, permed etc.). It has been proven that FAEEs are (surprisingly) not significantly affected by such treatments (Hartwig et al., 2003a). So far no systematic investigations in this regard have been carried out for EtG.

FAEE concentrations in hair from other body sites can be interpreted in a similar fashion as scalp hair (Hartwig *et al.*, 2003b). EtG: no information available.

Extensive studies involving over 1000 donors have been carried out since 2000. These have enabled us to establish reliable reference ranges for FAEEs with respect to drinking habits of the various groups:

Non to Moderate drinkers: < 0.4 ng/mg
Excessive/Abusive drinkers: > 1ng/mg

There are no reliable reference ranges for EtG from comprehensive studies. Further investigations are in progress to examine the applicability of the method in practice of the detection of alcohol abuse.

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