

Detection of EthylGlucuronide and EthylSulfate in urine after acute administration of different doses of alcohol

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Abstract

The present study aims to determine concentrations of EthylGlucuronide (EtG) and EthylSulfate (EtS) in urine after acute administration of different doses of alcohol. A single blind clinical trial was conducted in 53 healthy volunteers (32 males and 21 females), with previous drunk experiences. Subjects were distributed to 4 different doses: 20 g, 40 g, 60 g and 80 g of ethanol. Study variables included blood ethanol (BAL) and ethanol metabolites (EtG and EtS) concentrations in urine. Urine samples spiked with deuterated analogs as internal standards (EtG-d5 and EtS-d5) were analysed by LC-MS/MS following a dilute and shoot approach. Both, EtG and EtS urine excretion increased with the administered dose during the first 6 hours; EtS: from 63.95 μmol in 20 gr until 337.53 μmol in 80 gr; EtG: from 63.67 μmol in 20 gr until 405.38 μmol in 80 gr, in a non-linear manner. The excretion of EtG and EtS progressively decreased for all the alcohol doses and after 48 hours, excretion was below 20 μmol . After 72 hours, metabolites were undetectable.

Introduction

Alcohol use disorder is a relapsing disease associated with high levels of mortality and morbidity [1]. The amount of alcohol consumed has been related with the probability of comorbid diseases [1]. Ethanol metabolism occurs principally by oxidation to acetaldehyde and acetate by alcohol dehydrogenase and aldehyde dehydrogenase in the liver. Other minor pathways contributing to ethanol metabolic clearance are: catalase, CYP2E1 and the non-oxidative pathways [2]. The main non-oxidative metabolites of ethanol are: ethyl glucuronide (EtG), ethyl sulfate (EtS), ethyl nitrite (EtN), ethyl phosphate (EtP), phosphatidylethanol (PEth) and fatty acid ethyl esters (FAEE). Non-oxidative metabolites have been proposed as biomarkers of ethanol exposure due to its longer half-lives and are useful in the detection of relapses [3].

Aim

In the present study we aim to determine the dose-response of EthylGlucuronide (EtG) and EthylSulfate (EtS) formation in urine after acute alcohol administration to healthy volunteers.

Methods

A single blind clinical trial was conducted in 53 healthy volunteers with previous drunk experiences. Subjects were distributed to 4 different doses: 20 g, 40 g, 60 g and 80 g of ethanol. Study variables included blood ethanol (BAL) concentrations and ethanol metabolites (EtG and EtS) concentrations in urine.

To detect the EtG and EtS concentrations, urine samples were spiked with deuterated analogs as internal standards (EtG-d5 and EtS-d5) and were analysed by LC-MS/MS following a dilute and shoot approach.

Results

The final sample included 53 healthy volunteers (32 males and 21 females). The mean weight was 71.84 Kg and 61.54 Kg for men and women respectively and the BMI were 22.7 kg/m² and 22.67 kg/m² respectively.

As expected, ethanol concentrations increased in a dose-dependent manner (Figure 1). Both, EtG and EtS urine excretion also increased with the administered dose during the first 6 hours; EtS: from 63.9 μmol in 20 gr to up to 337.5 μmol in 80 gr; EtG: from 63.7 μmol in 20 gr to up to 405.4 μmol in 80 gr in a non-linear manner. The recovery of EtG and EtS progressively decreased for all the alcohol doses and after 48 hours, excretion was below 20 μmol . After 72 hours, metabolites were deemed undetectable (Figures 2 and 3).

Figure 1. Mean differences from basal blood concentrations of ethanol (mg/dL) depending on the dose administered

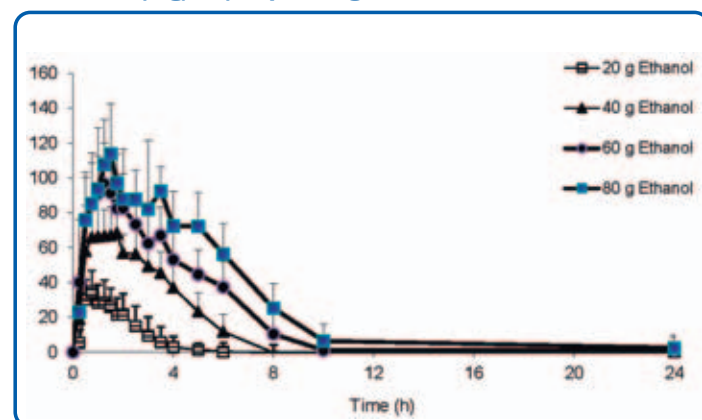


Figure 2. Recovery of EtG (μmol) in urine at different doses of ethanol

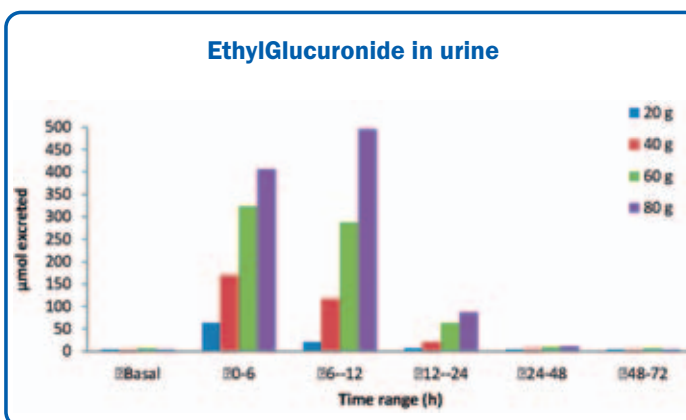
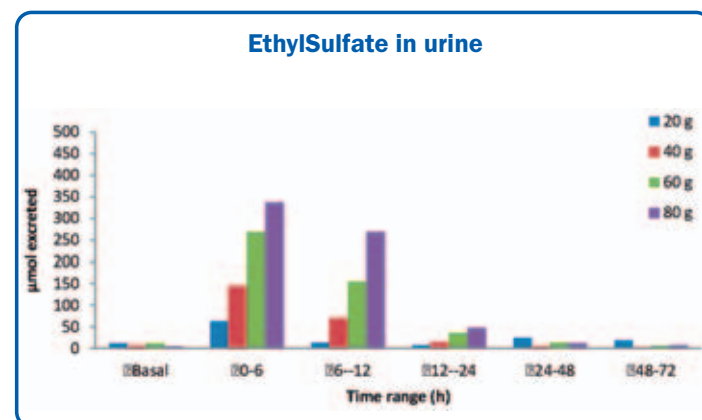


Figure 3. Recovery of EtS (μmol) in urine at different doses of ethanol



Conclusions

- EtG and EtS urine excretion after acute ethanol administration increases in a non-linear manner, with the dose.
- Metabolites were no longer detected 48 hours after alcohol acute administration.
- Studies in daily alcohol consumers should be performed in order to better understand the elimination of non-oxidative alcohol metabolites.

References

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