Impact of a Single Dose of a Probiotic Nutritional Supplement (AB001) on Absorption of Ethylalcohol: Results From a Randomized Double-Blind Crossover Study

Nutrition and Metabolic Insights
Volume 16: 1–6
© The Author(s) 2023
Article reuse guidelines:
sagepub.com/journals-permissions
DOI: 10.1177/11786388221141174

Andreas Pfützner[1,2], Anke Pfützner[3], Mina Hanna[1], Filiz Demircik[3], Daniela Sachsenheimer[1], Tobias Wittig[3] and Johan de Faire[4]

[1]Pfützner Science & Health Institute, Mainz, Germany. [2]Institute for Internal Medicine and Laboratory Medicine, University for Digital Technology in Medicine and Dentistry, Wiltz, Luxembourg. [3]Sciema GmbH, Mainz, Germany. [4]de Faire Medical AB, Stockholm, Sweden.

ABSTRACT

BACKGROUND: We conducted a prospective placebo-controlled double-blind randomized Study to assess the impact of a single dose of a nutritional Supplement (AB001) on alcohol absorption in healthy subjects. Other objectives were the impact on breath alcohol content, cognitive function 1 hour after alcohol uptake and tolerability.

METHOD: A total of 24 healthy volunteers were enrolled into the study (12 male, 12 female, age: 28.3 ± 10.8 years, BMI: 23.5 ± 5.7 kg/m^2). On the experimental day, they ingested a light breakfast together with a single dose (2 capsules) of AB001 (or placebo) and drank 2 moderate glasses of spirit (a total of 0.6 g/kg body weight). Breath alcohol tests and blood draws for determination of blood alcohol levels were performed for up to 6 hours. After crossover, the experiment was repeated in the following week. Areas under the curves were calculated to determine alcohol absorption rates.

RESULTS: There was a significant reduction of blood alcohol by 10.1% ($P < .001$) with AB001, when compared to placebo. There was a less pronounced but also significant reduction of alcohol in the breath test by 7.2% ($P < .05$). No difference in the cognitive function test between AB001 and placebo could be observed 60 minutes after alcohol ingestion (22.6 ± 8.0 seconds vs 23.0 ± 11.2 seconds, n.s.). The supplement uptake was well tolerated and there were no adverse events related to the study intervention.

CONCLUSION: Uptake of a single dose of AB001 shortly before drinking alcohol significantly reduced plasma alcohol and breath alcohol concentrations, but the effect was less pronounced compared to chronic uptake as shown previously.

RECEIVED: March 17, 2022. **ACCEPTED:** November 7, 2022.

TYPE: Original Research

FUNDING: The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This study was funded by de Faire Medical AB.

DECLARATION OF CONFLICTING INTERESTS: The author(s) declared the following potential conflicts of interest with respect to the research, authorship, and/or publication of this article: JD is founder and shareholder of de Faire Medical AB. APF has received a study grant and is consultant to de Faire Medical AB. The other authors have no conflict of interest to report.

CORRESPONDING AUTHOR: Andreas Pfützner, Pfützner Science & Health Institute, Haifa-Allee 20, Mainz D-55128, Germany. Email: andreas.pfuetzner@pfuetzner-mainz.com

Introduction

The fermentation of sugar into ethanol is one of the earliest biotechnologies employed by humans. The intoxicating effects of ethanol consumption have been known since ancient times. Ethanol has been used by humans since prehistory as the intoxicating ingredient of alcoholic beverages. Dried residue on 9000-year-old pottery found in China suggests that Neolithic people consumed alcoholic beverages. Today, social drinking is tolerated in many cultures around the world. It is accepted as a legitimate way to celebrate special occasions or just to relax after a hard day at work.[1,2] Drinking in moderation tends to be viewed as a harmless activity. It is only those who are habitually intoxicated who get judged as engaging in dangerous behavior. In reality, there is no level of alcohol use that can be considered completely risk free.[3,4] There are a number of benefits that people obtain from social drinking. This is why the activity has been popular for thousands of years. Alcohol is often described as a social lubricant. People tend to feel more relaxed after a drink or 2 and a bit less self-conscious. There are many social occasions that are based around alcohol consumption. Some studies even suggest that drinking in moderation may bring certain health benefits.[5-7] It is these beneficial aspects of alcohol that ensure its continued popularity.

However, even those people who drink in moderation can still encounter alcohol related problems. Alcohol is a toxin that can cause damage to the body even in small doses.[8,9] Those who drink regularly above the safe limits are at increased risk of health problems, including but not limited to certain cancers, cardiovascular events, high blood pressure, accidents while under the influence, and progression to alcohol abuse and addiction. The medical and social problems that are caused by the prolonged excessive consumption of ethanol in humans, span a spectrum of severity that can include impaired decision making, interpersonal problems, and serious physical consequences[10] as ethanol can have toxic effects on the liver, brain,[11] and heart,[12] among numerous organ systems. Acute alcohol intoxication, also known as drunkenness or alcohol poisoning, is the negative behavior and physical effect due to the recent

Creative Commons Non Commercial CC BY-NC: This article is distributed under the terms of the Creative Commons Attribution-NonCommercial 4.0 License (https://creativecommons.org/licenses/by-nc/4.0/) which permits non-commercial use, reproduction and distribution of the work without further permission provided the original work is attributed as specified on the SAGE and Open Access pages (https://us.sagepub.com/en-us/nam/open-access-at-sage).

drinking of alcohol. Symptoms at lower doses may include mild sedation and poor coordination. At higher doses, there may be slurred speech, trouble walking, and vomiting. Extreme doses may result in a respiratory depression, coma, or death. Acute alcohol intoxication accounts for the majority of alcohol-related disorders encountered in emergency rooms. Complications may include seizures, aspiration pneumonia, injuries including suicide, and low blood sugar.[13]

Alcohol is mostly metabolized in the liver, which is why the liver is particularly at risk of damage. Drinking heavily significantly increases the risk of alcoholic fatty liver, an early and reversible consequence of excessive alcohol intake. Chronic drinking alters the liver's metabolism of fats, and excess fat accumulates in the liver. Other effects on the liver include long-term inflammation (alcoholic hepatitis). This can lead to scar tissue and finally liver cirrhosis.[8,9,14]

de Faire Medical AB has developed a probiotic nutritional supplement (AB001), which is considered to help avoid such problems. Selected bacterial strains settle in the intestine tract where they preferably and effectively metabolize ethyl alcohol into CO_2 and water (Pfützner et al[15] and deFaire Medical, data on file). In consequence, less alcohol is expected to be absorbed by the body, and damage of organs through alcohol degradation products is expected to be diminished.

In a previous study, a substantial reduction of alcohol absorption into the blood by more than 70% was observed after 1 week of AB001 supplementation as compared to placebo, when drinking 0.3 g/kg of alcohol after a light breakfast. The reduction of measurable alcohol in the breath was also reduced significantly but to a lower extent as compared to the blood alcohol levels (by ~30%).[15] The purpose of this follow-up study was to answer the questions, whether a single dose of AB001, when taken directly prior to drinking alcohol, can also have a direct effect on alcohol absorption.

Subjects and Methods

The study was conducted in accordance with international and local ethical and scientific standards. The protocol was approved by the responsible ethical review board (Landesärztekammer Rheinland-Pfalz, Mainz, Germany) on December 23rd, 2020 and announced to the responsible national authority (Bundesamt für Verbraucherschutz und Lebensmittelsicherheit) also on December 23rd, 2020. Prior to participation the participants signed written informed consent. Thereafter, blood was drawn for the safety analysis and to identify potential exclusion criteria and the randomization into the 2 study arms took place (visit 0).

The study compound AB001 consists of naturally fermented rice bran containing Bacillus subtilis, B. coagulans, L-Cysteine and Dextrin (excipients: magnesium stearate salts, calcium phosphate and potassium phosphate) Contents/dose (800 mg): fermented rice bran 560 mg, L-Cysteine 200 mg, dextrin 4 mg and excipients 36 mg. The placebo formulation contained 800 mg of rice flour. The verum and placebo interventions were both provided in the same type of HPMC capsules and were visually indistinguishable from each other (Manufacturer: Optipharma AS, Drobak, Sweden, Batch numbers: 1912-09 (placebo), 00017014 (verum), expiry date Sept. 30th, 2022). Both interventions were stored at room temperature and were provided to the site with an expiry date after 12 months at the start of the study. To ensure double-blind study conduct, the randomization as well as the hand-out of the study interventions was performed by a staff member of the supervising contract research organization.

To be eligible for the study, subjects had to be healthy and of caucasian ethnicity and between 18 and 65 years of age. They needed to be willing to perform the 2 drinking experiments and had to have negative rapid tests for drug screening, alcohol consumption, and Covid-19 infection prior to the experiments. Main exclusion criteria were: any acute or chronic disease, presence or history of alcohol addiction, known allergy against probiotic nutritional supplements, regular uptake of any medication or dietary supplement, and pregnancy or breast feeding.

It is known from preclinical experiments that the bacteria provided with AB001 stay in the intestine tract for maximally 36 hours (data on file, de Faire Medical). Therefore, twice this period was considered as the minimal period between the experimental visits to allow for sufficient wash-out of the previous intervention.

The enrolled subjects were asked to participate in 2 experimental procedures (visits 1 and 2). The subjects came to the site in the morning after an overnight fast of at least 8 hours. In addition, they were asked to refrain from drinking alcohol and also from consuming probiotic substances on the 3 days before each of the experimental visits. After arrival at the study site in the morning, they were randomized to receive either placebo or the AB001 supplement. One hour after ingesting the investigational product the participant drank a first shot of a high alcoholic spirit (vodka; 0.3 g/kg body weight, timepoint 0 minute) and consumed a light breakfast with rolls, ham or jam, and with tea or coffee. The amount of alcohol to be administered in this trial was discussed and agreed upon with the IRB. Thereafter, they drank a second glass of alcohol (vodka; 0.3 g/kg bodyweight, timepoint 30 minutes). Blood was drawn for determination of plasma alcohol concentrations and breathalyzer assessments were be made for determination of breath alcohol concentrations after 0, 15, 30, 45, 60, 75, 90, 120, 180, 240, 300, 360, and 420 minutes.

Plasma alcohol concentrations were determined by an external laboratory (MVZ Augsburg, Germany) by means of a photometric method (CoBas 6000, c 501 module, Roche, Basel, Switzerland, lower limit of detection: 0.01‰ (=promille)). Breath alcohol concentrations were determined by the site staff using a point-of-care electrochemical sensor (Alcotest 3820, Dräger, Lübeck, Germany, lower limit of detection: 0.05‰).

The participants stayed at the study site for the whole time. A standardized lunch was served after 4 hours. In addition, the

participants performed a cognitive function test (number connection test, NCT-A[16]) at time-points 0 and 60 minutes. Information regarding adverse events was documented. If no alcohol was detectable in the breath at 2 consecutive timepoints after the second alcohol uptake, the experiment was prematurely terminated. The subjects stayed under medical supervision at the site for the entire time of the experimental visits to detect any possible adverse event. They were also asked to contact the site in case of any adverse event in the following 3 days after the experiment.

Number connection tests (NCT) have been used for many decades to assess mental performance.[16] The NCT measures cognitive processing speed and is involving psychomotor responding. The participant received a sheet of paper with 25 numbers and was asked to draw a continuous line from number 1 to number 25, connecting all numbers in between without elevating the pencil from the paper. The time was recorded in seconds. In case of a mistake, the investigator/study personnel asked the participant to correct the mistake without stopping the time.

The second experiment was performed 3 to 5 days later following the same experimental protocol. After the second experiment (visit 2), the subjects were discharged from the study.

The data was evaluated using methods of standard exploratory and descriptive statistical analyses to gain an understanding of the qualitative and quantitative nature of the collected data. For quantitative variables, arithmetic means, medians, standard deviations, and minimum and maximum values were calculated. Appropriate parametric and non-parametric statistical tests were used to compare the collected results. An F-test for detection of potential carryover effects was conducted, when appropriate, and Student's *t*-test was used to compare the areas under the curve of the blood and breath alcohol concentrations measured during the 2 exposure experiments. A *P*-value $< .05$ was considered to be statistically significant.

Results

The planned number of 24 healthy subjects were enrolled into the study (12 men, 12 women, mean age: 28.3 ± 10.8 years (range: 20-56 years), BMI: 23.5 ± 5.7 kg/m^2 (range: 16.9-31.1 kg/m^2). All of them performed the study per protocol and were included into the safety and efficacy analysis.

The amount of ingested alcohol (2×0.3 g/kg body weight) resulted in measurable blood alcohol levels in all subjects. The highest observed alcohol concentrations were both seen in the placebo experiments with 0.88‰ (breath) and 0.91‰ (blood), respectively (AB001: 0.77‰ and 0.72‰). A reduction in alcohol absorption by more than 5% was seen in 54% of the participants, while no change (±5%) was seen in 25% (breath: 54%/17%). The individual patient results are provided in Table 1.

The mean blood concentrations after nutritional supplementation with 2 capsules of study product or placebo 1 hour before the experiment are provided in Figure 1.

The $AUC_{Blood(0\text{-}420\ min)}$ was calculated to be 116 ± 32‰*min from the placebo experiments and 104 ± 24‰*min from the AB001 experiments (−10.1%; $P < .05$).

The mean breath alcohol concentrations after nutritional supplementation with 2 capsules/day of study product or placebo 1 hour before the experiment are provided in Figure 2.

The $AUC_{Breath(0\text{-}420min)}$ as calculated from the mean concentrations was 98 ± 29‰*minutes from the placebo experiments and 91 ± 25‰*minutes from the AB001 experiments (−7.2%, $P < .05$).

The uptake of the ingested amount of alcohol did not impact the cognitive function of the subjects as assessed by measuring the time required to complete the standardized number connection test. There was a minor impairment in the performance of the individual subject after uptake of the alcohol, which was statistically significant in the AB001 experiment (baseline: 19.8 ± 7.8 seconds vs 22.6 ± 8.0 seconds, $P < .05$) and almost significant in the placebo experiment (19.5 ± 5.7 seconds vs 23.0 ± 11.2 seconds, $P = .057$).

The uptake of the nutritional supplement and placebo was well tolerated. Only 2 minor adverse events were reported, which were both mild in nature and classified to be not related to the study interventions (mild headache and mild anemia detected in the screening laboratory results).

Discussion

There is a large body of evidence in the literature that probiotics are effective for the prevention and treatment of alcoholic liver disease. Alcohol disrupts the composition of intestinal flora; probiotics modulate the gut microbiota and reverse alcohol-associated intestinal barrier dysfunction by decreasing intestinal mucosal permeability and preventing intestinal bacteria from translocating. Probiotics are living microorganisms that play beneficial roles, for example, by supporting the maintenance the balance of intestinal microbiota.[17] The physiological functions of probiotics are achieved directly or indirectly by adjusting the composition of the host intestinal microbiota, activating the endogenous microbial community and regulating the immune system.[18] Known mechanisms by which probiotics may help to prevent or improve alcoholic liver injury include an increase antioxidant activity leading to reduction in oxidative stress and promotion of antioxidant production,[19,20] improvement of alcohol-induced lipid metabolism[21] and treatment of alcoholic fatty liver,[22,23] reduction of inflammatory cytokine expression in liver and intestine,[24,25] a modulation of the mucosal immune system leading to an enhanced intestinal immune barrier function,[26–28] and generally a beneficial regulation of the intestinal flora.[29] It has also been suggested that restoration of microbial metabolites is another logical treatment approach. Supplementation with probiotics may increase protective cytokine and hormonal expression by modulating the gut microbiota; and bioengineered bacteria can deliver therapeutics to the microbiota or host. It has also been

Table 1. Individual subject alcohol absorption (AUC for blood and breath alcohol) after single uptake of AB001 or placebo.

SUBJECT NO.	AGE [YEARS]	GENDER	BMI [KG/M²]	BLOOD ALCOHOL $AUC_{0\text{-}420MIN}$ [‰*MIN]		BREATH ALCOHOL $AUC_{0\text{-}420MIN}$ [‰*MIN]	
				PLACEBO	AB001	PLACEBO	AB001
1	23	F	23.7	80	59	70	44
2	23	F	24.2	149	117	126	95
3	32	M	19.1	73	71	61	59
4	25	F	23.9	132	151	126	139
5	29	M	23.0	110	109	93	87
6	27	F	20.8	87	105	83	100
7	56	F	25.1	147	122	132	105
8	20	M	20.4	87	89	74	83
9	30	M	31.1	210	151	181	153
10	21	M	21.8	128	105	99	98
11	23	F	16.9	95	107	70	98
12	22	F	23.5	135	111	122	102
13	31	M	25.3	115	106	83	89
14	26	F	25.7	113	115	99	106
15	55	M	28.9	79	88	62	69
16	27	M	26.8	115	106	110	98
17	20	F	21.7	148	83	95	69
18	28	F	18.9	81	75	63	56
19	22	M	23.2	111	82	90	80
20	23	F	21.6	130	139	123	126
21	26	M	26.8	115	113	96	88
22	33	F	27.6	157	129	131	103
23	36	M	24.1	94	87	82	74
24	21	M	20.1	82	76	81	65
Mean	28	12 M	23.5	116	104	98	91
STD	11	12 F	5.7	32	24	29	25

suggested that drugs can be used to change and modulate bacterial enzymes or pathways.[30,31]

To our knowledge, AB001 is the first probiotic supplement that has been demonstrated to substantially reduce alcohol absorption in the intestine tract. A composition of specifically selected bacterial strains directly metabolizes ethyl alcohol into CO_2 and water. In consequence, ethyl alcohol is degraded prior to further resorption through the intestine mucosa, does not reach the blood, and cannot induce negative metabolic activities and organ damage anymore. The purpose of our clinical investigations was to understand the degree of effectiveness of this protective approach and, if possible, to elucidate different application regimens for this new probiotic supplement with respect to safety and tolerability.

In a first randomized placebo-controlled double-blind crossover study,[15] 24 healthy subjects (13 male, 11 female, age: 25.4 ± 7.7 years, BMI: $23.6 \pm 2.5\,kg/m^2$) were randomized to take 2 capsules/day of AB001 or placebo for 1 week prior to an alcohol exposure experiment. On the experimental day, they ingested a light breakfast and drank a moderate glass of spirit (0.3 g/kg body weight). A significant reduction of blood alcohol levels by 70.3% ($P < .005$ vs placebo) was seen with AB001, (breath test: −30.7%; $P < .005$ vs placebo). No difference was seen in a cognitive function test performed 60 minutes after

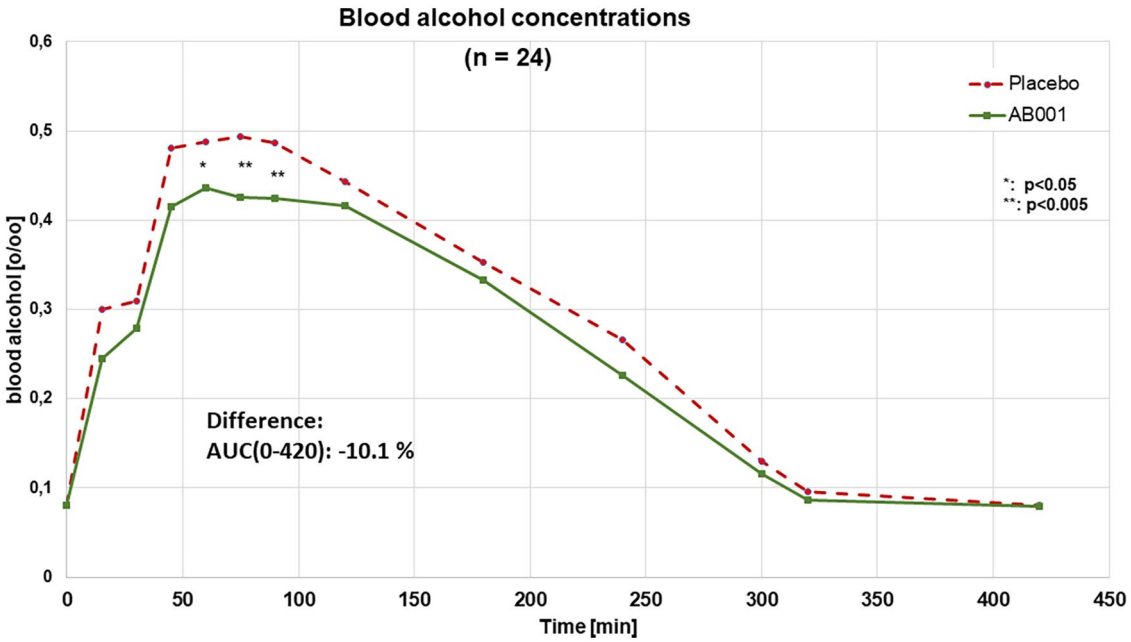

Figure 1. Mean blood alcohol concentrations after uptake of 2 × 0.3 g/kg body weight with a light breakfast in between (30 minutes between alcohol uptakes, n = 24).

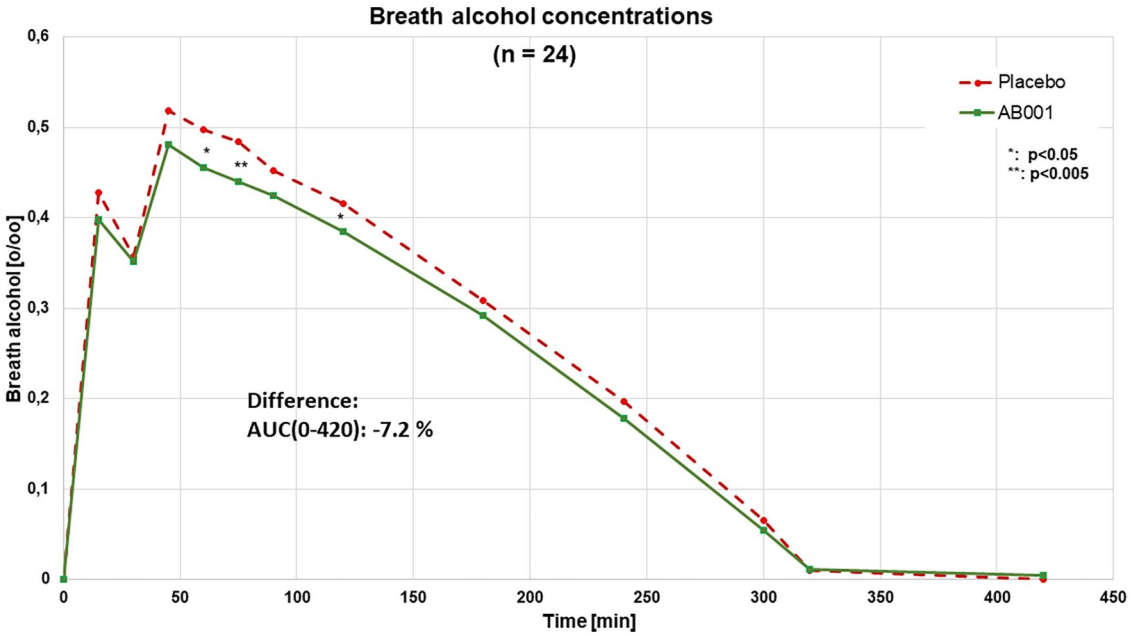

Figure 2. Mean breath alcohol concentrations after uptake of 2 × 0.3 g/kg body weight with a light breakfast in between (30 minutes between alcohol uptakes, n = 24).

alcohol ingestion (22.4 ± 7.7 seconds vs 22.7 ± 5.6 seconds, n.s.). There were no adverse events or serious adverse events reported in the previous study.[15]

In this current trial, it was observed that uptake of a single dose of AB001 from the same manufacturing lot than in the first study taken only 1 hour prior to drinking 2 × 0.3 g/kg of alcohol with 30 minutes time interval and uptake of a light meal between the 2 shots did not lead to similar reduction of alcohol absorption as seen in the previous trial. While, the AUC of the alcohol concentrations in the blood was significantly reduced by about 10% and breath alcohol was significantly reduced by 7% with AB001 as compared to placebo, it remains open, whether this represents already a clinically relevant reduction. In contrast to the first study, we had doubled the amount of alcohol and measurable alcohol concentrations in blood and breath were now seen with all participants most likely because the first shot of alcohol was taken prior to the meal. One explanation for the observed lower efficacy is that uptake of the single dose of

AB001 1 hour before drinking may not have given the bacterial strains sufficient time to properly settle into the intestine tract to enable significant bacterial metabolism of the alcohol prior to absorption. Although it is known that Bacillus spores under optimal conditions can germ within 5 to 10 minutes,[32] the conditions in the intestine tract may not be good enough to see full efficacy with 1 dose just taken 1 hour prior to drinking alcohol. Based on the results of both studies, it is tempting to speculate that AB001 can develop its protective effect on the liver better, when it is taken on a regular basis, for example, 2 capsules per days for at least 2 to 3 days.

The study has several limitations which need to be considered when interpreting the results. Firstly, the number of subjects is quite small, and the study hence only has an exploratory pilot character. Secondly, the intervention was only a single dose taken 1 hour prior to drinking, while the previous study data and individual anecdotal experience is suggesting that the probiotic supplement should be chronically administered to see a higher liver protective efficacy. Our actual recommendation for an occasional drinking situation, which is derived from uncontrolled observations and personal experience, is to take at least 2 doses prior to start drinking. The first dose should preferably be taken in the morning of that day and a second dose should be taken at least 2 hours prior to drinking. Another limitation is that the current study like the previous trial was conducted in the morning after an overnight fast, while alcohol is usually consumed later in the day and in the evening. Any diurnal effect of the supplement and differences in its effect on alcohol absorption in the evening cannot be ruled out at this point. Larger and more comprehensive prospective and confirmatory studies will be required before drawing general conclusions about the effectiveness and tolerability of the probiotic nutritional supplement.

In conclusion, uptake of a single dose of AB001, a probiotic supplement developed to prevent alcohol absorption in the intestine tract, within 1 hour prior to starting to drink alcohol resulted in a significant decrease in blood alcohol and breath alcohol concentrations, which was less pronounced compared to the impact of a more chronic supplementation as shown in a previous trial.

Author Contributions

Planing of the study design and preparation of the protocol (AP1, AP2, FD, TW, JdF), submission to IRB and the authorities (AP1, AP2, FD, TW), study conduct and data generation (AP1, MH, DS), data analysis (AP1, MH, DS, JdF), and preparation and review of the manuscript (all).

Supplemental Material

Supplemental material for this article is available online.

REFERENCES

1. Khaderi SA. Introduction: alcohol and alcoholism. *Clin Liver Dis*. 2019;23:1-10.
2. Stettler A. Alcohol—historical aspects. *Ther Umsch*. 1990;47:344-349.
3. Lindgren KP, Baldwin SA, Olin CC, et al. Evaluating within-person change in implicit measures of alcohol associations: increases in alcohol associations predict increases in drinking risk and vice versa. *Alcohol Alcohol*. 2018;53:386-393.
4. Cummins J, Lindgren KP, DeHouwer J. On the role of (implicit) drinking self-identity in alcohol use and problematic drinking: a comparison of five measures. *Psychol Addict Behav*. 2021;35:458-471.
5. Ronksley PE, Brien SE, Turner BJ, Mukamal KJ, Ghali WA. Association of alcohol consumption with selected cardiovascular disease outcomes: a systematic review and meta-analysis. *BMJ*. 2011;342:d671.
6. Di Castelnuovo A, Costanzo S, Bagnardi V, Donati MB, Iacoviello L, de Gaetano G. Alcohol dosing and total mortality in men and women: an updated meta-analysis of 34 prospective studies. *Arch Intern Med*. 2006;166:2437-2445.
7. Howard AA, Arnsten JH, Gourevitch MN. Effect of alcohol consumption on diabetes mellitus: a systematic review. *Ann Intern Med*. 2004;140:211-219.
8. Zakhari S. Overview: how is alcohol metabolized by the body? *Alcohol Res. Health*. 2006;29:245-254.
9. Lieber CS, DeCarli LM, Feinman L, et al. Effect of chronic alcohol consumption on ethanol and acetaldehyde metabolism. *Adv Exp Med Biol*. 1975;59:185-227.
10. Rehm J, Taylor B, Mohapatra S, et al. Alcohol as a risk factor for liver cirrhosis: a systematic review and meta-analysis. *Drug Alcohol Rev*. 2010;29:437-445.
11. Pascual M, Blanco AM, Cauli O, Miñarro J, Guerri C. Intermittent ethanol exposure induces inflammatory brain damage and causes long-term behavioural alterations in adolescent rats. *Eur J Neurosci*. 2007;25:541-550.
12. Urbano-Marquez A, Estruch R, Navarro-Lopez F, Grau JM, Mont L, Rubin E. The effects of alcoholism on skeletal and cardiac muscle. *N Engl J Med*. 1989;320:409-415.
13. Howard RJ, Slesinger PA, Davies DL, Das J, Trudell JR, Harris RA. Alcohol-binding sites in distinct brain proteins: the quest for atomic level resolution. *Alcohol Clin Exp Res*. 2011;35:1561-1573.
14. Parker R. The natural history of alcohol-related liver disease. *Curr Opin Gastroenterol*. 2020;36:164-168.
15. Pfützner A, Hanna M, Andor Y, et al., eds. Chronic uptake of a probiotic nutritional supplement (AB001) inhibits absorption of ethylalcohol in the intestine tract—results from a randomized double-blind crossover study. *Nutr Metab Insights*. 2022. doi.
16. Lezak MD, Howieson DB, Loring DW. (eds.) *Neuropsychological Assessment*. 4th ed. Oxford University Press; 2004.
17. Lambert JC, Zhou Z, Wang L, Song Z, McClain CJ, Kang YJ. Prevention of alterations in intestinal permeability is involved in zinc inhibition of acute ethanol-induced liver damage in mice. *J Pharmacol Exp Ther*. 2003;305:880-886.
18. Lata J, Jurankova J, Kopacova M, Vitek P. Probiotics in hepatology. *World J Gastroenterol*. 2011;17:2890-2896.
19. Wang Y, Kirpich I, Liu Y, et al. Lactobacillus rhamnosus GG treatment potentiates intestinal hypoxia-inducible factor, promotes intestinal integrity and ameliorates alcohol-induced liver injury. *Am J Pathol*. 2011;179:2866-2875.
20. Wang Y, Liu Y, Sidhu A, Ma Z, McClain C, Feng W. Lactobacillus rhamnosus GG culture supernatant ameliorates acute alcohol-induced intestinal permeability and liver injury. *Am J Physiol Gastrointest Liver Physiol*. 2012;303:G32-G41.
21. Zhang M, Wang C, Wang C, et al. Enhanced AMPK phosphorylation contributes to the beneficial effects of Lactobacillus rhamnosus GG supernatant on chronic-alcohol-induced fatty liver disease. *J Nutr Biochem*. 2015;26:337-344.
22. Koutnikova H, Genser B, Monteiro-Sepulveda M, et al. Impact of bacterial probiotics on obesity, diabetes and non-alcoholic fatty liver disease related variables: a systematic review and meta-analysis of randomised controlled trials. *BMJ Open*. 2019;9:e017995.
23. Bajaj JS. Alcohol, liver disease and the gut microbiota. *Nat Rev Gastroenterol Hepatol*. 2019;16:235-246.
24. Wang Y, Liu Y, Kirpich I, et al. Lactobacillus rhamnosus GG reduces hepatic TNFα production and inflammation in chronic alcohol-induced liver injury. *J Nutr Biochem*. 2013;24:1609-1615.
25. Bajaj JS, Heuman DM, Hylemon PB, et al. Randomized clinical trial: lactobacillus GG modulates gut microbiome, metabolome and endotoxemia in patients with cirrhosis. *Aliment Pharmacol Ther*. 2014;39:1113-1125.
26. Shukla PK, Meena AS, Manda B, et al. Lactobacillus plantarum prevents and mitigates alcohol-induced disruption of colonic epithelial tight junctions, endotoxemia, and liver damage by an EGF receptor-dependent mechanism. *FASEB J*. Published online June 18, 2018. doi:10.1096/fj.201800351R
27. Yan F, Liu L, Dempsey PJ, et al. A Lactobacillus rhamnosus GG-derived soluble protein, p40, stimulates ligand release from intestinal epithelial cells to transactivate epidermal growth factor receptor. *J Biol Chem*. 2013;288:30742-30751.
28. Zhao H, Zhao C, Dong Y, et al. Inhibition of miR122a by Lactobacillus rhamnosus GG culture supernatant increases intestinal occludin expression and protects mice from alcoholic liver disease. *Toxicol Lett*. 2015;234:194-200.
29. Hendrikx T, Duan Y, Wang Y, et al. Bacteria engineered to produce IL-22 in intestine induce expression of REG3G to reduce ethanol-induced liver disease in mice. *Gut*. 2019;68:1504-1515.
30. Sarin SK, Pande A, Schnabl B. Microbiome as a therapeutic target in alcohol-related liver disease. *J Hepatol*. 2019;70:260-272.
31. Gu Z, Liu Y, Hu S, et al. Probiotics for alleviating alcoholic liver injury. *Gastroenterol Res Pract*. 2019;2019:9097276.
32. Vary JC, Halvorson HO. Kinetics of germination of *Bacillus* spores. *J Bacteriol*. 1965;89:1340-1347.