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Cooking with beer: How much alcohol is left?

Scientific Paper

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Abstract

When cooking with beer and other alcoholic beverages the loss of ethanol relative to loss of water determines the final concentration of ethanol in the food, but predicting the rate of loss is not simple. Since many people for various reasons (drivers, pregnant women etc.) may strictly want to limit their ethanol intake, it is important to obtain knowledge on this topic. Knowing the final ethanol concentration in prepared foods is also crucial for precisely calculating the energy content of a food. In the current study ethanol was quantified using gas chromatography in ten foods prepared with beer: vinaigrette, pancake, carrot soup, rye bread porridge, steamed fish, spareribs, braised beef, rye bread and wheat bread before, during and after preparation. The estimated amount of ethanol per serving was calculated accordingly. The final concentrations in the foods were in the range from 2.62% (v/v) and 2.48% (w/w) to below detection limit. The highest estimated amount of ethanol per serving was accordingly 1.28 g which would be of little concern to most people. Theoretical concentration values calculated from the recipe were in most cases higher than the measured ones, since these values do not reflect the loss during preparation. Nor do the theoretical concentration was higher than the theoretical. The heat-treated foods as demonstrated by the rye bread in which case the measured ethanol concentration was higher than the theoretical. The heat-treated foods generally decreased in ethanol concentration during preparation, implying that a higher proportion of the initial amount of ethanol has been lost than of water. The decrease in ethanol concentration observed during cooking further implies that the cook can control the final ethanol content of a food by adjusting cooking time. The other parameter in control of the cook is the initial concentration as prescribed by the recipe.

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Introduction

Alcoholic beverages are used as an ingredient in a variety of foods across cultures, not least in Denmark where beer traditionally has been used in staple foods like rye bread

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porridge (Jensen, 1953; Nimb, 1900; Strunge and Strunge, 1924). An extensive cooking literature focuses on traditional as well as innovative food preparations with beer or wine as an ingredient (Botelet, 2008; Ellis, 1975; La France, 1997; Waldo, 1958). The use of alcoholic beverages in cooking has the purpose of adding flavour to the food but it may also change the texture of foods. One very common use is as cooking liquid in meaty dishes or as an ingredient in a sauce.

Relatively few scientific studies have explored the use of wine and beer in cooking. The changes in flavour when cooking with wine have been studied by Rognså (2014) and Snitkjær et al. (2011) which both showed a loss of many

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volatile aroma compounds during cooking making the aroma of the wines more alike after cooking.

The health effect of using beer and wine marinades in food preparation has been studied by Gorelik et al. (2008), Melo et al. (2008) and Viegas et al. (2012, 2014) who showed a reduction in the levels of carcinogenic compounds in meat when marinated in wine and beer prior to frying.

One concern when cooking with alcoholic beverages is the concentration of ethanol in the cooked dish. This is important when serving the food to pregnant women, children, drivers and to anyone who wants to control their alcohol intake strictly. The amount of retained ethanol during food preparation is also relevant for energy calculations of a meal since ethanol lost during cooking should not be counted as energy in the final serving. A few studies have been published on the topic (Augustin et al., 1992; Hansen et al., 2012; Helander and Bergström, 2001; Mateus et al., 2011) giving some insight on ethanol loss during cooking. Broader knowledge is however valuable in order to predict and not least control the remaining amount of ethanol relative to water when cooking with beer and other alcoholic beverages.

The current study provides knowledge on ethanol concentrations in ten types of foods and the corresponding amount of ethanol per serving. The study further provides knowledge on the change in ethanol concentration during cooking. The foods chosen comprise a range of different heat treatments and are prepared with different beers. The data provided and the following discussion on the general principles for ethanol loss upon cooking are intended to be used as a guideline for predicting the ethanol concentration in foods prepared with alcoholic beverages. The provided knowledge aims to help control one's alcohol intake and support correct energy calculations in foods prepared with alcoholic beverages.

Materials and methods

Ten foods were prepared according to Table 1. All recipes originate from cookbooks but were in some cases modified slightly in order to fit into the study. Details on sampling times are also given in Table 1. In the case of steamed fish and braised beef samples of the meat/fish and the liquid were taken separately. Samples were frozen at -18 °C prior to ethanol analyses.

Ethanol concentration was determined using static headspace gas chromatography mass spectrometry (HS-GC–MS). Each analysis was performed in duplicate.

For liquid samples, 1 ml of sample was mixed with 9 ml water in a 20 ml headspace vial and 50 μ L of internal standard (9.99% methanol) were added. The ethanol concentrations of these foods are given as a volume percentage (also known as ABV).

Solid samples were weighed out for headspace analysis and results given as a weight percentage (also known as ABW). Samples of 15g were weighed into a 250 ml centrifuge tube and 135g water was added. The mixture was homogenised using a polytron type PT 10–35 homogeniser. Ten ml homogenised samples were weighed into a 20 ml headspace vial and 50 µL internal standard (9.99% methanol) were added.

Quantification of ethanol was done by creating matrixcalibration curves for each food. This was done by spiking the foods prepared without alcoholic beverage (using water instead). In case of the pancakes, rye bread, wheat bread, braised beef and spareribs, separate standard curves were made for the food before and after heat treatment. The bread was a special case because the'blind sample' prepared, substituting beer with water, contained ethanol due to the fermentation process. So for the dough and bread the standard addition method was used, with one non-spiked and 6 spiked levels.

Gas chromatographic analyses were carried out on a Trace GC Ultra gas chromatograph with a split/splitless injection port coupled to a DSQ quadrupole mass spectrometer (Thermo). Headspace sampling was carried out using a CTC CombiPAL sampler (CTC Analytics AG). Headspace sampling was performed after incubation at 60 °C for 10 or 25 min. 250 μ L of the headspace was sampled using a 2.5 ml syringe thermostated at 90 °C. Samples were injected in splitless mode (3 min), and injection port temperature was 250 °C.

Separation of compounds (ethanol and methanol) was done using a CP-WAX 52 capillary column (50 m x 0.32 mm internal diameter, 0.45 μ m film thickness, Agilent). Helium with a constant flow of 1 ml/min was used as a carrier gas. After injection the column was kept at 40 °C for 10 min, and then raised at 30 °C/min to 240 °C. The temperature of the transfer line connected to the mass spectrometer was set at 260 °C.

Detection was performed using a mass spectrometer in electron-impact (EI) ionisation mode with electron energy of 70 eV. Quantifications were performed in full-scan mode, mass range m/z 15–300, with a scan rate of 1.6863 scans/s. Total ion chromatogram (ethanol) and m/z 32 (methanol) were used for quantifications.

Results

A variety of foods were prepared with beer according to recipes. The foods were selected to represent a variety of heat treatments and ingredients. The ethanol concentration measured in the prepared foods, presented in Table 2, range from not detectable (ND) to 2.62% (v/v) and 2.48% (w/w). Ethanol concentrations in foods must be evaluated in relation to the amount of the food normally consumed within a meal. Serving sizes are estimated according to Ygil (2013) and the corresponding amount of ethanol per serving is presented in Table 2. The highest value of 'ethanol per serving' is 1.28 g, which corresponds to no more than 11% of a lager type beer (330 ml, 4.6% v/v), which contains approximately 12 g ethanol.

In addition to the measured ethanol concentrations, theoretical ethanol concentrations have been calculated based on the recipe, without considering the loss during preparation, see Table 2. These theoretical ethanol concentrations are as expected generally higher than the measured concentrations in the final products. Rye bread and vinaigrette are exceptions.

Among the foods prepared with beer, vinaigrette was the only one not heat treated. The concentration of ethanol in the

Table 1

Recipe of ten foods prepared for the study. References to the original recipe are given (except for wheat bread); preparation of the dishes is slightly modified from the original recipe in order to fit the experimental design. Blank samples for the gas chromatographic determination of ethanol were prepared by substituting beer for water in the recipe.

Food (ref.)	Ingredients	Quant. batch	Preparation and sampling All ingredients were mixed with a hand held blender. Vinaigrette was prepared in triplicate. Samples were taken of the two-phase liquid right after blending.		
Vinaigrette (Evald, 2005)	Jacobsen Velvet Ale, 5.9% (Carlsberg) Olive oil (Extra Virgin, Santagata) Lime juice Mint leaves Zittauer onions Garlic Sugar (Dansukker) Salt (Coarse salt, Salina) Water	100 g 50 ml 8 g 3 pc 40 g 3 g 4.5 g 3 g 20 g			
Pancakes (Ritterband, 1992)	Carlsberg Pilsner 4.6% Egg (Danæg) Milk (Arla, 1.5% fat) Butter (Xtra) Wheat flour (COOP Änglemark) Sugar (Dansukker) Salt (Salina)	50 g 3 400 ml 100 g 125 g 30 g 2 g	Wet and dry ingredients were mixed separately and then everything was mixed together. Melted butter was added at the end of mixing. 70 ml dou was used for each pancake. Pancakes were baked on a preheated pan (EV trio, non-stick, diameter: 17.5 cm) 1 min on each side on medium/high he Samples were taken from the dough and the pancakes. Three replicate batc were made.		
Rye bread porridge (Evald, 2005)	Gamle Carlsberg Porter 7.8% (Carlsberg) Water Danish rye bread (KerneKlaus, COOP) Icing sugar (Dansukker)	150 g 200 ml 150 g 16 g	Bread was soaked in water for 2 h and then blended with beer and sugar. The bread mixture was cooked in an Eva trio steal pan, diameter: 16 cm. It was brought to the boil at high heat while stirring (3 min) and then cooked at low heat while stirring for 8 min. Samples were taken from the uncooked bread-beer mixture (time 0), just as it started to boil (time 3 min) and after 6 and 11 min of heating respectively. Two replicate batches were made.		
Carrot soup (Ritterband, 1992)	Carlsberg Pilsner 4.6% Vegetable stock concentrate (Bong) Water Carrots (Søris), peeled and sliced Milk (Arla, 1.5% fat) Wheat flour (COOP Änglemark) Butter (Xtra)	330 g 27 ml 573 ml 300 g 100 ml 10 g 30 g	Carrots were cooked in water and stock concentrate until tender, (3 min at high heat until boil $+12$ min at low heat). Butter was melted in a separate po flour added and then milk. When boiling, beer was added and cooked for another minute after which the carrots, including their cooking liquid, were added. The soup was boiled for another 0, 2 or 10 min prior to blending an sampling. Two replicate batches of the soup was prepared for each of the three cooking times: 0, 2 and 10 min. The soup was prepared in EVA trio ster pot, diameter: 21 cm.		
Steamed fish (Evald, 2005)	Jacobsen Velvet Ale, 5.9% (Carlsberg) Cod (Thorfisk) Red bell pepper, chopped Green bell pepper, chopped Zittauer onions (Gyldensten), chopped Celery sticks Potato, cut in halves Bay leaf Cumin Sunflower oil (First Price) Salt (Salina)	400 g 600 g 320 g 320 g 130 g 210 g 600 g 4 pc 2 g 24 g 8 g	Onions were sautéed in oil. Additional vegetables were added. Beer was added and the mixture brought to the boil at high heat. Fish was put on the top of the steaming vegetables and steamed in pot (Eva trio steal, diameter: 28 cm) for 30 min at low heat after which samples of both liquid and fish were taken. Two replicate batches were made.		
Spare ribs (Bogø and Stig, 2005)	Carlsberg Pilsner 4.6%	300 g			

Table 1	(continued)
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Food (ref.)	Ingredients	Quant. batch	Preparation and sampling		
	Beef back ribs (Danish Crown) Garlic, crushed Zittauer onions (Gyldensten), chopped Conc. tomato puree (Petti) White wine vinegar (Monari Federzoni) Thyme Honey (Budget) Worchestersauce (Lea & Perrins) Mustard, whole grain (Maile) Ketchup (Beauvais)	3200 g 12 g 230 g 180 g 140 g 1 g 80 g 100 g 13 g 140 g	All ingredients for the marinade were well mixed. Meat was precooked in the pot by starting in cold water and bringing the water to the boil. The meat was drained and water discarded. After cooling the marinade was applied and the marinated meat rested in the fridge $(-5 ^{\circ}\text{C})$ for 24 h. Meat was roasted in hot air oven (Rational) first for 15 min at 210 $^{\circ}\text{C}$ and then 75 min at 150 $^{\circ}\text{C}$. Samples were taken during roasting at time 0 (just before entering the oven), 30 min and 90 min. Two replicate batches were prepared.		
Braised beef (Evald, 2005)	Jacobsen Original Dark Lager, 5.8% (Carlsberg) or Red wine, 14.5% (Chile, 2011) ^a Beef fore shank Oil sunflower Carrots (Søris, organic), chopped Zittauer onions (Gyldensten), chopped	500 g 1000 g 24 g 150 g 120 g	Meat was pre-roasted at 300° C in hot air oven (Rational) with oil for 10 min. Additional ingredients were added to the roasting pan which was covered with a lid and the dish was braised at 150 °C for 180 min in hot air oven (Rational). Samples were taken both from meat and liquid at time 0 (just after all ingredients had been added), and after 180 min of braising. Additionally liquid samples were taken at 60 and 120 min. Two replicate batches were prepared.		
Wheat bread yeast fermentation	Jacobsen Brown Ale, 6% (Carlsberg) Wheat flour Water Fresh yeast (long/ short fermentation) Salt (Salina) Sugar (Dansukker)	350 g 900 g ^b 300 ml 5/50 g 20 g 7 g	Yeast was dissolved in liquid. Additional ingredients were added. Dough was kneaded by machine (Kenwood) for 5 min and leavened at room temperature till double size (20 h/1.5 h). Portions of 200 g dough were placed in silicone baking cups. Baked at 190 $^{\circ}$ C for 20 min in hot air oven (Rational). Samples were taken of the dough before and after leavening, and from the baked bread. Two replicate batches were made.		
Wheat bread sourdough ferm.	Jacobsen Brown Ale, 6% (Carlsberg) Wheat flour (COOP) Water Sour dough (Meyers) Salt (Salina) Sugar (Dansukker)	350 g 900 g 250 ml 150 g 20 g 7 g	Same procedure as described for the yeast fermented wheat bread, using so dough instead of yeast. Leavening for 20 h at room temperature. Two replica batches were made.		
Rye bread (Bogø and Stig, 2005)	Gamle Carlsberg Porter, 7.8% Buttermilk (Arla) Rye flour, coarse (COOP) Wheat flour (COOP Änglemark) Salt (Salina) Fresh yeast (De Danske Gærfabrikker) Water Apple (Joya), grated Dark brown sugar (Muscova, Dansukker)	600 g 150 ml 950 g 300 g 27.5 g 10 g 400 ml 200 ml 10 g	<i>Day 1</i> : Buttermilk was mixed with 100 g rye flour, 5.5 g salt and 10 g yeast. Dough was leavened for 17 h at 22 °C. <u>Day2</u> : Water, 400 ml porter, 450 g rye flour, wheat flour, apple, 11 g salt and sugar was added to the soured dough and kneaded by hand. Dough was leavened 17 h at 22° C. <u>Day 3</u> : 400 g rye flour, 200 ml porter and 11 g salt was added to dough and it was kneaded by hand. 1300 g of dough was placed in a 1 L tin and leavened for 90 min at 20 °C. Baked for 60 min in a preheated hot air oven (Bosch) 170 °C. Samples were taken on day 3 only, from the leavened dough and from the baked bread. Two replicate batches were made.		

^aMade from Cabernet Sauvignon, Camenere, Shiraz; Palo Alto, Maule Valley.

^bBlank sample was made with 30% more flour in order to give it the same consistency.

Table 2

Theoretical and measured concentrations of ethanol (\pm standard deviations) in various foods. ND = Not detectable. The theoretical values are calculated based on the amounts given in the recipe, the labelled ethanol concentration in the beverage and estimated densities of ingredients. Liquid foods are quantified as a volume percentage (v/v) and solid samples as a weight percentage (w/w) and the numbers are given as mean \pm standard deviation (n=2 or 3, depending on dish). Serving sizes are estimated based on report by Ygil (2013) and the amount of ethanol per serving determined accordingly. Wheat breads (WB) differ in way they are leavened: yeast-slow (20 h using 0.6% yeast based on flour weight), yeast-fast (1.5 h using 5.6% yeast based on flour weight), and SD-slow (20 h using sourdough).

Food	Ethanol concentration (%)		Serving size	Ethanol per serving (g)	LOD/LOQ ^d	Conc. Unit %
	Theoretical	Measured				
Vinaigrette	2.26	2.62 ± 0.18	15 ml	0.31	0.01/0.03	v/v
Pancake						
Pancake dough	0.24	0.28 ± 0.02	_	_	0.02/0.05	v/v
Pancake		0.11 ± 0.03	50 g ^a	0.06	0.01/0.04	w/w
Rye bread porridge	1.87	0.40 ± 0.00	250 g	1.00	0.01/0.04	w/w
Carrot soup	1.39	0.65 ± 0.06	250 ml	1.28	0.03/0.10	
Steamed fish						
Liquid	5.9	0.80 ± 0.09	75 ml	1.09	0.02/0.05	v/v
Fish meat	0	0.49 ± 0.06	125 g		0.03/0.08	w/w
Spare ribs	0.25	ND	100 g	-	0.03/0.08	w/w
Braised beef (BB)						
BB - meat (wine)	0	0.08 ± 0.04	100 g	0.21	0.03/0.08	w/w
BB - liquid (wine)	13.5	0.22 ± 0.00	75 ml		0.02/0.05	v/v
BB - meat (beer)	0	0.06 ± 0.01	100 g	0.07	0.004/0.01	w/w
BB - liquid (beer)	5.8	< 0.02	75 ml		0.003/0.02	v/v
BB - meat (beer), prior to cooking	0	0.04 ± 0.02	-	-	0.004/0.01	w/w
Wheat bread (WB)					LOQ	
WB: yeast-slow	1.04	0.85 ± 0.07	45 g ^b	0.38	< 0.05	w/w
WB: yeast-fast	1.01	0.80 ± 0.07	45 g^{b}	0.36	< 0.05	w/w
WB: SD-slow	0.98	0.59 ± 0.07	45 g ^b	0.27	< 0.05	w/w
WB water: yeast-fast	0	0.10°	45 g ^b	0.05	< 0.05	w/w
Rye bread (RB)					LOQ	
RB (beer)	1.41	2.48 ± 0.12	45 g ^b	1.12	< 0.05	w/w
RB (water)	0	1.21 ^c	45 g^{b}	0.54	< 0.05	w/w
RB (water) - bakery		0.24 ^c	45 g ^b	0.11	< 0.05	w/w
RB (water) - supermarket		0.34 ^c	45 g ^b	0.15	< 0.05	w/w

^aOne serving corresponds to one pancake.

^bOne serving corresponds to one slice.

^cOnly one replicate available.

 d LOD/LOQ are based on the signal to noise approach; i.e. SN=3:1 for LOD and SN=10:1 for LOQ. For bread samples, LOQ is estimated based on SN of the unspiked sample, relative to the concentration of this sample determined by the standard addition procedure.

vinaigrette was 2.62% (v/v), as shown in Table 1, which is generally high compared to the cooked foods. Using a serving size of 15 ml vinaigrette one would consume 0.31 g ethanol in one serving. The theoretical ethanol concentration calculated from the list of ingredients is 2.26% (v/v). The difference between theoretical and measured concentrations may be the result of inaccuracy in calculating the theoretical value since some inaccuracy of mass calculations of the ingredients and the labelled ethanol concentration on the beer is expected. A decrease in ethanol concentration over time may have occurred if the vinaigrette had been kept either in fridge or at room temperature, particularly if stored in an open container, but this was not investigated in the current study. In a similar study, Augustin et al. (1992) reports a decrease in ethanol concentration from 2.06% to 1.77% (w/w) of a brandy pie during cold overnight storage in an uncovered container.

Pancakes are known in many food cultures, although the recipes may vary considerably with respect to the choice of liquid, flour type and leavening agent. In Denmark beer has traditionally been added to pancake doughs (Strunge and Strunge, 1924). As shown in Table 2 the pancake dough prepared with beer contained 0.28% (v/v) ethanol, which resembles the theoretical value, while the finished pancake (baked 2 min on the pan) contained only 0.11% (w/w) ethanol. This concentration is lower than many fermented breads prepared without alcohol as evaluated by Logan and Distefano (1998). The ethanol intake from consuming these pancakes is correspondingly very small, 0.06 g per pancake, assuming a pancake size of 50 g.

The rye bread porridge (in Danish: ' \emptyset *llebrød*', which literally means 'beer bread') is a very traditional dish in Denmark made from (stale) rye bread, beer and sugar and

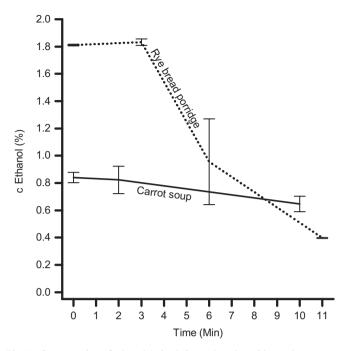


Fig. 1. Concentration of ethanol (w/w%) in rye bread porridge and concentration of ethanol (v/v%) in carrot soup as a function of cooking time. The figure shows mean data with standard deviations (n=2). The porridge was brought to the boil within the first 3 min of cooking after which a sharp decrease in ethanol concentration is seen. In the carrot soup beer was added to the boiling soup at time 0.

served with cream. The bread is usually soaked in beer and then cooked in beer and/or water. In our experiment, the ethanol concentration in the uncooked rye bread porridge was 1.81% (w/w) which decreased to 0.40% (w/w) during the 11 min of cooking, see Fig. 1. The theoretical concentration calculated is 1.87% (w/w) which is close to the measured initial concentration, but much higher than the actual concentration in the cooked porridge. The concentration in the cooked porridge corresponds to an intake of 1.00 g ethanol when consuming one serving of 250 g, see Table 2. As seen from Fig. 1, the decrease in ethanol concentration was steep from 3 min onwards, at which time the porridge reached the boil. The steep decrease with time indicates that the cooking time is an important factor for the final concentration of ethanol; in case of only 5 min cooking, the concentration would have been approximately 1.2% (w/w) and the ethanol intake correspondingly higher.

The carrot soup had a similar preparation time, 10 min, although in this case beer was added to the boiling soup. The first sample (corresponding to time 0) was taken after adding the last ingredients at which time the beer had already been in the pot for one minute. Results, presented in Fig. 1, show a slight decrease in ethanol concentration during cooking, from the initial 0.84% (v/v) to the final concentration of 0.65% (v/v). The relatively small decrease in ethanol concentration due to a considerable loss of ethanol in the first minute after adding the beer, prior to sampling. This assumption is supported by the theoretical ethanol concentration, which is 1.39% (v/v), much higher than the first measured concentration (time 0).

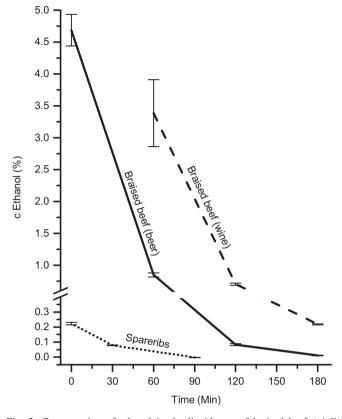


Fig. 2. Concentration of ethanol in the liquid part of braised beef (v/v%) prepared with wine or beer and in spare ribs (w/w%) as a function of cooking time in the oven. The figure shows mean data with standard deviations (n=2). The concentration axis is broken from 0.3% to 0.6%. Data on braised beef (liquid) prepared with wine at time 0 is missing, but the liquid consisted of pure red wine with an alcohol concentration of 13.5 v/v% according to the label. Ethanol values that are below LOD are shown as '0' in the figure and those below LOQ are shown as the LOQ value.

Assuming a serving size of 250 ml one serving contains 1.28 g ethanol, which is comparable to the porridge and the steamed fish.

The steamed fish was prepared by steaming fish and vegetables for 30 min in beer. As seen from Table 2, the resulting ethanol concentration was 0.80% (v/v) in the liquid part and 0.49% (w/w) in the fish meat. The liquid was initially pure beer with an alcohol concentration of 5.9% (v/v) according to the label. During steaming the beer was diluted with juice from the vegetables and the fish and ethanol evaporated, which caused the ethanol concentration to drop considerably. Assuming that the fish did not contain any ethanol before cooking, it has apparently absorbed some during the steaming. Consuming a serving size of 125 g fish plus 75 ml liquid one would get 1.09 g ethanol plus possibly a little from the vegetables, not included here.

The spare ribs demonstrate another use of beer in cooking. In this case the meat was marinated in a beer-containing marinade prior to roasting. As seen from Fig. 2 and Table 2 the ethanol concentration was low, 0.22% (w/w), to begin with and ethanol was not detectable after 90 min in the oven. The theoretical concentration was 0.25% (w/w), resembling the initial value. Studies have shown a reduction in the level of

carcinogenic compounds when marinating meat in beer or wine prior to frying (Gorelik et al., 2008; Melo et al., 2008; Viegas et al., 2012, 2014). Based on the current study, one can marinate meat in beer without the risk of alcohol intake when applying a long heat treatment. Mateus et al. (2011) studied the ethanol concentrations in steak marinated in beer or wine and grilled eight minutes. They report a concentration in grilled steak of 0.153% (w/w) when marinated in wine and 0.08% (w/w) when marinated in beer. Using wine for the marinade combined with a much shorter heating time apparently resulted in a considerably higher ethanol concentration in the grilled steak. One has to consider both the amount and choice of alcoholic beverage used but also the heating applied.

The braised beef, braised in the oven for 3 h, represents the most intense of the heat treatments applied in this study. The dish was prepared with both beer and wine in order to study effect of the choice of beverage on the ethanol concentration in the dish. The ethanol was quantified in the meat and the liquid separately. The beer-braised meat contained 0.06% (w/w) ethanol and the wine-braised meat 0.08% (w/w) ethanol as shown in Table 2. The meat used for beerbraising contained 0.04% (w/w) ethanol before preparation and thus no significant absorption of ethanol took place in the meat during braising. The meat part of the dish consequently contributes very little to the alcohol intake whether wine or beer is used as braising liquid. Fig. 2 shows how the ethanol concentration in the liquid part of the braised beef decreases as a function of cooking time. The liquid consisted of pure wine or beer to begin with and the ethanol concentration just before entering the oven (time 0) was accordingly high. There is, however, a dramatic decrease in ethanol concentration during the cooking time. As shown in Table 2, the final concentrations in the liquid after 3 h of cooking are 0.22% (v/v) for wine and below 0.02% (v/v) for beer. Fig. 2 also shows that the choice of wine versus beer matters more in the case of a shorter braising time, and presumably less with a longer braising time. After just 1 h of braising, the ethanol concentration in the liquid part of the beer-braised dish has dropped to just below 1% (v/v) whereas it takes 2 h for the ethanol concentration to reach this level in the liquid when using wine for braising. In the study by Mateus et al. (2011) ethanol concentrations were measured in three comparable dishes, fish stew, beef bib and hunter rabbit, that had simmered 45-60 min. The concentrations reported were in the range from 0.02% to 0.67% (w/w) and depended on the dish and the amount of wine added.

Assuming a serving size of 100 g meat plus 75 ml liquid the intake would be 0.21 g when braised with wine and 0.07 g when braised with beer (see Table 2). This value is comparable to the meat dishes prepared and evaluated by Augustin et al. (1992) who report 0.2 g per serving in case of 'Pot roast Milano' which simmered with wine for $2\frac{1}{2}$ h and 0.4–0.6 g in 'orange chicken burgundy' simmered in wine for 10 min. Augustin et al. (1992) moreover report values in the range of 1–3 g ethanol per serving in the additional dishes that were studied (scalloped oyster, Grand Marnier sauce, flamed cherry jubilee and brandy pie).

Wheat bread is a staple in many Western food cultures. Water and dairy products are the common liquid ingredients; recipes with beer are however also widespread. Purified yeast and/or sour dough are common leavening ingredients. Yeast added either as purified yeast or as part of a sourdough contributes to the ethanol concentration in breads since ethanol is produced during yeast fermentation. In the current study, three wheat breads (WB) were prepared with beer using three leavening methods, varying in leavening agent used (yeast 0.6%/veast 5.6%/sourdough) and leavening time (1.5 or 20 h). The results, presented in Table 2, show that the ethanol concentrations range from 0.59% (w/w) when using sourdough to 0.85% (w/w) using yeast. For comparison ethanol concentration in plain wheat bread reported by Grosch and Schieberle (1991) are 0.39% (w/w) in crumb and 0.18% (w/w) in crust. In the study by Logan and Distefano (1998) ethanol concentrations in a variety of industrial baked products are reported in the range from 0.03% to 1.662% (w/w), of which the baked goods with the higher concentrations (>0.5%) all contain fruit, onions or brandy.

The higher ethanol concentration when using pure yeast as compared to sourdough in the current study is supposedly caused by a higher ethanol contribution from the yeast fermentation when using pure yeast as compared to using sourdough that contains acetic acid and lactic acid bacteria as well as yeast. This can be studied further in Fig. 3, which shows how the ethanol concentration develops during the preparation steps: unleavened dough, leavened dough and bread. The ethanol concentrations in the three unleavened doughs made with beer are similar, ranging from 0.95% (w/w) when prepared with sourdough to 0.99% (w/w) when prepared with yeast (slow), which is slightly lower than the calculated theoretical concentrations in the dough ranging from 0.98% to 1.04% (w/w). After leavening the slowly yeast-fermented dough reaches the highest ethanol concentration, 1.52% (w/ w), followed by the fast leavened yeast dough and the sourdough-fermented dough. These observations show that both the added beer and the fermentation process contribute to the final concentration of ethanol and further show that the choice of leavening method influences the amount of ethanol present in the bread. The production of ethanol during yeast dough fermentation has been studied by Jayaram et al. (2013). They measured the ethanol content in wheat doughs as a function of fermentation time (0-180 min) with yeast concentrations ranging from 1.0% to 5.3% (w/w, based on flour weight) at 30 °C. They clearly demonstrate that ethanol concentration in the dough increases with higher yeast concentrations and longer fermentation time. In the dough containing 5.3% yeast (comparable to the fast fermented dough in the current study) ethanol concentrations increased from 0 to approximately 1.6% (w/w) in 3 h. These findings indicate that the fermentation process produces a considerable amount of ethanol in yeast-raised breads and that this is important to consider when evaluating the alcohol intake in relation to bread consumption.

Fig. 3 further shows that ethanol concentrations decrease in the three wheat doughs during baking (20 min) but remain in

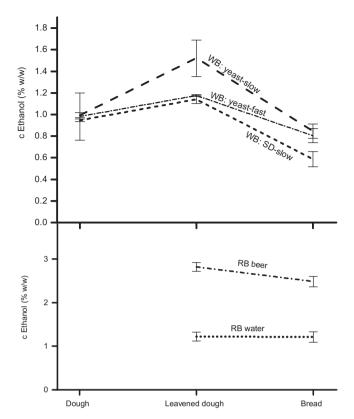


Fig. 3. Concentration of ethanol (w/w %) in three wheat breads (WB) and two rye breads (RB) and in the corresponding dough, before and after leavening. The figure shows mean data with standard deviations (n=2). The wheat breads were prepared with beer with one exception (WB water) and are defined by the leavening agent used and the leavening time: yeast-slow (20 h using 0.6% yeast based on flour weight), yeast-fast (1.5 h using 5.6% yeast based on flour weight) and SD-slow (20 h using sourdough). Rye bread was prepared with beer according to recipe and with water, substituting beer for water in same recipe. In case of the rye breads there is no data on ethanol levels in dough before leavening.

the same order with respect to the final ethanol concentrations. The decrease in ethanol concentration during baking is explained by evaporation. The evaporation of ethanol during baking may, however, to some extent be counteracted by the continuous production of ethanol during baking, which can take place until the elevating temperature is sufficient to inactivate the yeast cells. Based on the presented results a slice (45 g) of bread prepared with beer contains 0.27–0.38 g ethanol.

Bread made from rye is a staple in Denmark and many recipes prescribe the use of beer. Rye bread is traditionally made with sourdough and a correspondingly long fermentation time. In the present experiment a rye bread was made according to a recipe with beer. Ethanol concentrations were measured in the leavened dough and in the bread. As shown in Fig. 3, the ethanol concentration in the leavened dough was 2.82% (w/w) when made with beer and 1.22% (w/w) when made with water (blank sample), demonstrating that the high concentration in the beer dough is caused partly by the added beer and partly by the fermentation during the long leavening time ($35\frac{1}{2}h$ in total). The theoretical concentration values for the rye bread are 1.41% (w/w) with beer and 0% without beer

(presented in Table 2), which in both cases are considerably lower than the concentrations measured in the leavened dough, demonstrating the increase in ethanol concentration during leavening, where fermentation takes place.

From Fig. 3 it can also be seen that the ethanol concentration drops from 2.82% to 2.48% (w/w) during baking of the dough with beer whereas the concentration of the dough with water maintains almost the same value after baking. In both cases the drop in ethanol concentration during baking of the rve breads for 60 min is smaller than the case of the wheat breads presented above in the same figure, baked for only 20 min. Whether this is the result of a more efficient retention of the ethanol in the rye bread and/or due to more efficient yeast fermentation during the baking is uncertain. The resulting homemade rye breads contain 2.48% (w/w) ethanol when made with beer and 1.21% (w/w) ethanol when prepared with water, which is a surprisingly high concentration compared to the theoretical values (1.41% and 0% w/w, with and without beer, respectively) but also relatively high compared to the results of the wheat breads presented above and the values reported in the literature (Logan and Distefano, 1998). For comparison ordinary plain rye bread (without beer) was purchased in a bakery and a supermarket. The ethanol concentrations in the purchased rye breads, as shown in Table 2, were somewhat lower, 0.24–0.34% (w/w). These values resemble the level reported by Logan and Distefano (1998), who found a concentration of 0.33% (w/w) in a commercial sourdough rye bread. When consuming the homemade rye bread one would get 1.12 g ethanol per slice of rye bread with beer and 0.54 g in case of the rye bread prepared without beer. In case of the purchased rye bread one would only get no more than 0.15 g per slice of bread. The most prominent explanation for the high concentrations in the homemade rye bread is the very long leavening time, giving time for the production of ethanol by yeast. There is apparently also a high retention during baking which is promoted by the baking tin and possibly also the crust formation on the bread.

Discussion

Since the estimated intake of ethanol per serving is maximum 1.28 g for the foods studied as compared to 12 g in a 4.6% (v/v) lager beer, the ethanol intake is considered to be of little concern to most people including children, pregnant women and drivers. It is however recommended to be careful if one decides to add alcoholic beverages to many components in a meal, since the total ethanol intake will add up. It should also be kept in mind that the concentration in a similar dish may be noticeably higher than reported here if the initial concentration is higher or the preparation technique is different. These results can therefore only be seen as a guideline for the chef and the consumer.

People with alcohol misuse and prescribed disulfiram drug are of a particular concern, since treatment with disulfiram drug causes a 'hangover' at small concentrations of ethanol in the blood. Mild effects may occur at blood alcohol concentrations of 5-10 mg/100 ml, a limit that in theory would be

exceeded if for example a woman weighing 50 kg consumes two gram alcohol instantaneously in which case her blood concentration can be expected to reach 7 mg/100 ml. Consequently, people taking disulfiram need to be very conscious about the alcohol concentration in foods, whether the alcohol originates from alcoholic beverages or from yeast fermentation during processing. It is however important to point out that the concentration in the blood stream is dependent on the rate of alcohol consumption, body weight of the consumer (Center for Substance Abuse Treatment, 2009) and type of beverage (Mitchell et al., 2014). It has additionally been shown that ingestion of food slows down the absorption of alcohol into the blood stream (Jones et al., 1997).

The measured ethanol concentrations in the prepared foods were generally lower than the initial concentration and lower than the initial concentration estimated based on the list of ingredients (theoretical concentration). These findings support the claim that energy calculations in a dish containing ethanol cannot be calculated correctly from the recipe alone. Since ethanol has a relatively high energy content, 29 kJ/g, precise information on ethanol concentrations is necessary in order to estimate the energy content of a meal correctly. In case of foods prepared with alcoholic beverages as in the current study, the energy content would often be overestimated if based on the recipe only.

The differences between the theoretical ethanol concentration values and the measured ones in the final prepared food product are generally high for the most intensively heated foods like the braised beef corresponding to the shown substantial decrease in ethanol concentration during preparation (see Fig. 2). It seems that the heating applied during preparation is at least to some degree associated with the decrease in ethanol concentration during preparation, as also claimed by Augustin et al. (1992). A deeper quantitative understanding of the effect of time and temperature on the loss of ethanol relative to water has however not been obtained.

With better knowledge on the final ethanol concentration in a range of foods prepared with alcoholic beverages, more basic scientific questions arise such as which parameters are essential for the retention of ethanol during cooking and how the resulting ethanol concentration can be controlled.

When using beer or any other alcoholic beverage for food preparations, the loss of ethanol will be caused by the volatility (as determined by Henry's law) of ethanol in the matrix; the volatility is dependent on temperature and increases with increasing temperature. Because of this temperature dependence a more severe loss of ethanol must be expected when temperatures are raised during heat preparation. The loss of water is however also dependent on temperature and water evaporation will also increase with increasing temperature. The change in ethanol concentration during cooking of foods containing both water and ethanol will depend on both water and ethanol loss. If ethanol concentration remains the same during food preparation the explanation may be that both ethanol and water are retained efficiently in the dish, or it may be that ethanol and water are lost at a rate that corresponds to the initial concentration. If ethanol on the other hand is lost at a rate that is higher than that of water (relative to the initial concentration), a decrease in ethanol concentration will occur. A decreasing ethanol concentration over time during cooking has been observed in the current study as well as in the two similar studies by Augustin et al. (1992) and Mateus et al. (2011). These studies as well as the present one do not state the loss of water upon cooking which would be useful in order to get a deeper understanding of the changes in ethanol concentration reported.

A fundamental understanding of ethanol loss during cooking can be hard to derive from studies on complex dishes. In a more simple study, Helander and Bergström (2001) quantified ethanol in mulled wine upon heating in an open pot. The result showed a decrease in ethanol concentration from the initial 14% (v/v) to 5% after five minutes. They further quantified ethanol in four water-ethanol mixtures with initial ethanol concentrations of 5%, 10%, 20% and 40% v/v, respectively, which had been cooked for five minutes in an open pot. These authors found that a higher initial concentration resulted in a higher final concentration although the final differences were smaller. They also reported that when heating the 20% waterethanol mixture the ethanol concentration decreased linearly over time during the first five minutes of cooking.

The initial concentration (the recipe) is one parameter that can be controlled in the kitchen. Choosing beer over wine will decrease the ethanol concentration but in many cases like for example the braised beef, the differences in the final dish are negligible. The choice of beer versus wine may easily have a much higher impact on the final flavour of the dish than on the ethanol concentration. Mateus et al. (2011) investigated the influence on initial ethanol concentration in two dishes, fish stew and beef bib, cooked for 35 and 60 min respectively. They found a significant higher concentration in the cooked dishes when increasing the initial concentration from 0.67-2.01% (w/w) in the fish stew and from 0.46-1.38% (w/w) in the beef bib. The concentrations were however all below 0.7%(w/w) and thus the differences would be of little importance to most people. Generally it can be said that in those foods where ethanol concentration changes only little during the preparation, the initial concentration will have a high impact on the final concentration, like the vinaigrette. In foods that decrease considerably in ethanol concentration during preparation, like the braised beer, the initial concentration is of less importance.

The cooking time is another parameter that has an impact on the final ethanol concentration of a food. In case of a very steep decrease in concentration with time, the cooking time matters very much for the final concentration, and less so in those foods were ethanol concentration is less dependent on cooking time. Cooking time can to some degree be controlled in the kitchen, but the chef will of course need to think about other quality parameters as well when deciding upon cooking time.

Based on the obtained results in the current study, many other questions arise related to which parameters affect the loss of ethanol during food preparation. How much impact does the temperature have on the change in ethanol concentration over time? Augustin et al. (1992) suggest that the 'severity' of the heat treatment determines the alcohol retention but the effect of temperature and time has not been systematically investigated. Does the choice of heating method make a difference, for example oven heating versus heating in a pot? Augustin et al. (1992) suggest that an oven baked dish retains alcohol more efficiently due to a lower rate of heat transfer in dry heat systems as compared to a more efficient rate of heat transfer in wet heat systems such as simmering. This has likewise not been investigated systematically.

How much impact does the food matrix have or the matrix of the surface of the food? Augustin et al. (1992) suggest that bread crumbs on their scalloped oyster dish have retained the ethanol efficiently during oven cooking. In the current study we found a surprisingly high ethanol concentration in the homemade rye bread which may at least partly be caused by an efficient retention of ethanol by the crust during baking.

Do the dimensions of the pot have any influence on the ethanol concentration? Augustin et al. (1992) suggest that a larger diameter in the pot leads to a sharper decrease in concentration based on their results although this has not been investigated thoroughly.

In conclusion the study has contributed to an increased knowledge on ethanol concentration in foods prepared with beer, the expected intake of ethanol per serving and how this may be controlled by the cook. A number of questions related to the fundamental understanding of ethanol loss during cooking arise from the study, which call for further studies in the field.

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References

- Augustin, J., Augustin, E., Cutrufelli, R.L., Hagen, S.R., Teitzel, C., 1992. Alcohol in food preparation. J. Am. Diet. Assoc. 92, 486–488.
- Bogø, J., Stig, H., 2005. Øl til. Forlaget d3, Copenhagen.
- Botelet, A., 2008. The Gourmet's Guide to Cooking with Wine. Quarry Books, Beverly, Massachusetts.
- Center for Substance Abuse Treatment, 2009. Incorporating Alcohol Pharmacotherapies into Medical Practice. Report (SMA) 09-4380. Substance Abuse and Mental Health sevices Administration (US), Rockville.
- Ellis, A., 1975. Wine Lovers Cookbook. Hutchinson & Co, London.
- A., Evald, 2005. Mad med øl. Gyldendal, Copenhagen.

- Gorelik, S., Ligumsky, M., Kohen, R., Kanner, J., 2008. Novel function of red wine polyphenols in humans: prevention of absorption of cytotoxic lipid peroxidation products. FASEB J. 22, 41–46.
- Grosch, W., Schieberle, P., 1991. Bread. In: Maarse, H. (Ed.), Volatile Compounds in Foods and beverages. Marchel Dekker, New York, pp. 41–77.
- Hansen, C.E., Kwasniewski, M.T., Sacks, G.L., 2012. Decoupling the effect of heating and flaming on chemical and sensory changes during flambé cooking. Int. J. Gastron. Food Sci. 1 (2), 90–95.
- Helander, A., Bergström, M., 2001. Kan man koka bort alkoholen ur glöggen?. Lakartidningen 98, 5554.
- Jayaram, V.B., Cuyvers, S., Lagrain, B., Verstrepen, K.J., Delcour, J.A., Courtin, C.M., 2013. Mapping of Saccharomyces cerevisiae metabolites in fermenting wheat straight-dough reveals succinic acid as pH-determining factor. Food Chem. 136 (2), 301–308.
- Jensen, K., 1953. Frøken Jensens kogebog. Gyldendal, Copenhagen.
- Jones, A.W., Jönsson, K.Å., Kechagias, S., 1997. Effect of high-fat, highprotein, and high-cabohydrate meals in the pharmacokinetics of a small dose of ethanol. Br. J. Clin. Pharmacol. 44, 521–526.
- La France, P., 1997. Cooking and eating with beer, 50 Chefs, brewmasters, and Restaurateurs Talk about Beer and Food. John Wiley & Sons, New York.
- Logan, B.K., Distefano, S., 1998. Ethanol content of various foods and soft drinks and their potential for interference with a breath-alcohol test. J. Anal. Toxicol. 22 (3), 181–183.
- Mateus, D., Ferreira, I.M.P.L.V.O., Pinho, O., 2011. Headspace SPME–GC/ MS evaluation of ethanol retention in cooked meals containing alcoholic drinks. Food Chem. 126 (3), 1387–1392.
- Melo, A., Viegas, O., Petisca, C., Pinho, O., Ferreira, I.M.P.L.V.O., 2008. Effect of beer/red wine marinades on the formation of heterocyclic aromatic amines in pan-fried beef. J. Agric. Food Chem. 56 (22), 10625–10632.
- Mitchell, M.C., Teigen, E.L., Ramchandani, V.A., 2014. Absorption and peak blood alcohol concentration after drinking beer, wine and spirits. Alcohol Clin. Exp. Res. 38 (5), 1200–1204.
- Nimb, L., 1900. Fru Nimbs kogebog. H. Hagerups forlag, Copenhagen.
- Ritterband, M., 1992. Ølkogebogen. Gyldendal, Copenhagen.
- Rognså, G.H., 2014. Emulsions from a Culinary Perspective The Case of Hollandaise Sauce and Its Derivatives (Ph.D. thesis). University of Copenhagen.
- Snitkjær, P., Risbo, J., Skibsted, L.H., Ebeler, S., Heymann, H., Harmon, K., Frøst, M.B., 2011. Beef stock reduction with red wine – Effects of preparation method and wine characteristics. Food Chem. 126 (1), 183–196.
- Strunge, M., Strunge, A., 1924. Koge- og bagebog for alle hjem. Mariane Strunges Forlag, Aarhus.
- Viegas, O., Amaro, L.F., Ferreira, I., Pinho, O., 2012. Inhibitory effect of antioxidant-rich marinades on the formation of heterocyclic aromatic amines in pan-fried beef. J. Agric. Food Chem. 60 (24), 6235–6240.
- Viegas, O., Yebra-Pimentel, I., Martínez-Carballo, E., Simal-Gandara, J., Ferreira, I.M.P.L.V.O., 2014. Effect of beer marinades on formation of polycyclic aromatic hydrocarbons in charcoal-grilled pork. J. Agric. Food Chem. 62 (12), 2638–2643.
- Waldo, M., 1958. Beer and Good Food: Brighten Your Menus and Recipes with Beer and Ale. Doubleday & Company, New York.
- Ygil, K.H., 2013. Mål, vægt og portionsstørrelser på fødevarer. Technical University of Denmark, Søborg.