On the Significant Influence of Water on the Formation Mechanisms of 5-Acetyl-3,4-dihydro-2*H*-1,4-thiazine

*W. ENGEL and P. SCHIEBERLE**

Deutsche Forschungsanstalt für Lebensmittelchemie, Garching bei München, Germany,
**E-mail: peter.schieberle@lrz.tu-muenchen.de*

Abstract: The formation of 5-acetyl-3,4-dihydro-2*H*-1,4-thiazine in Maillard-type reactions of fructose with cysteamine under dry heating and cooking conditions was studied. Labelling experiments with 2-^{13}C-fructose revealed, that the formation pathways are completely different, depending on the water content of the mixture. Under dry heating conditions, 5-(1-^{13}C-acetyl)-3,4-dihydro-2*H*-1,4-thiazine is formed almost exclusively with the 2-^{13}C of fructose found at the carbonyl carbon of the acetyl group. Under cooking conditions, ADHT is mostly unlabelled and most probably formed from erythrulose. Erythrulose might be generated from 2-^{13}C-fructose by loss of 1-^{13}C-acetic acid, indicated by the high amount of the latter found in the mixture. A possible mechanism leading from fructose to erythrulose is postulated.

Keywords: ADHT; 2-^{13}C-fructose; acetic acid

INTRODUCTION

The amino acid cysteine is well accepted in the literature as one of the most relevant precursors of very potent sulfur compounds formed during food processing. Depending on the reaction parameters, the degradation of cysteine may lead to quite different, very reactive intermediates, such as H_2S, cysteamine or mercaptoethanal.

Although cysteamine has been used as a source to generate aroma-active thiazolidines for use in chocolate, smoky and meat-like aromas [1, 2], only a few data are available on volatiles generated from a direct reaction of cysteamine with a carbohydrate. Recently, we have identified 5-acetyl-3,4-dihydro-2*H*-1,4-thiazine (ADHT) as one of the major aroma active compounds in Maillard-type reactions of fructose with cysteamine [3]. However, even though this potent compound was identified earlier [4], yet there is no reliable mechanistic data available on the formation pathways of this compound.

The aim of this study was, therefore to clarify the formation of ADHT under either simulated dry heating or simulated cooking conditions using fructose, labelled with ^{13}C at the carbonyl carbon atom.

EXPERIMENTAL

Model reactions. To simulate cooking conditions, cysteamine (0.23 mmol) was dissolved in phosphate buffer (7 ml, 0.1 mol/l, pH 7.0) and reacted with 2-^{13}C-fructose (0.7 mmol) in a laboratory autoclave by raising the temperature within 20 min from 20°C to 145°C. To mimic dry heating conditions, cysteamine (0.23 mmol) and 2-^{13}C-fructose (0.7 mmol) were intimately mixed with silica containing 10% water (w/w). The mixture was heated for 10 min. at 150°C in a metal block.

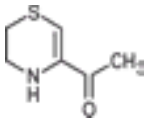
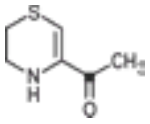
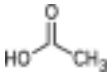
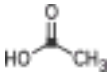
Analytical methods. ADHT was extracted from both model reactions using diethylether and analysed by HRGC-MS after SAFE-distillation under high vacuum. Mass spectra of ADHT and acetic acid were obtained in HR, EI and CI mode.

RESULTS AND DISCUSSION

Labelling distribution in ADHT and acetic acid

From the mass spectra (data not shown), the following distribution of the labelling was calculated (the ^{13}C atom is represented by the small black rectangle) – Table 1.

Table 1. Labelling distribution in ADHT and acetic acid under dry heating and cooking conditions

		
ADHT		
Cooking	76%	24%
Dry heating	5%	95%
		
Acetic Acid		
Cooking	38%	62%
Dry heating	85%	15%

Dry heating conditions

From the results it is very likely that under dry heating conditions nearly all of the ADHT is formed by a single pathway. However, this pathway does not seem to involve the classical Amadori product formation because the label is found nearly exclusively at the carbonyl-carbon of ADHT. The proposed mechanism based on the labelling experiment is depicted in Figure 1.

Cooking conditions

Under cooking conditions the major amount of ADHT is unlabelled and additionally, acetic acid is mostly and if then exclusively labelled at the carboxy-carbon atom, indicating, that acetic acid is removed from the fructose skeleton, leaving erythrulose as remainder. Erythrulose again, by virtue of its chemical structure, should be a perfect precursor for ADHT in combination with cysteamine. This was verified in a separate experiment using erythrulose and cysteamine generating large amounts of ADHT (data not shown). A reasonable mechanism deduced from these experiments is depicted in Figure 2.

CONCLUSIONS

The results of the labelling experiments clearly indicate that the pathways leading to ADHT are completely different, depending on the reaction conditions. Under dry heating conditions reactions seem to be favoured starting from the pyranose or furanose form of fructose by direct water elimination. It has to be mentioned, that the pathway

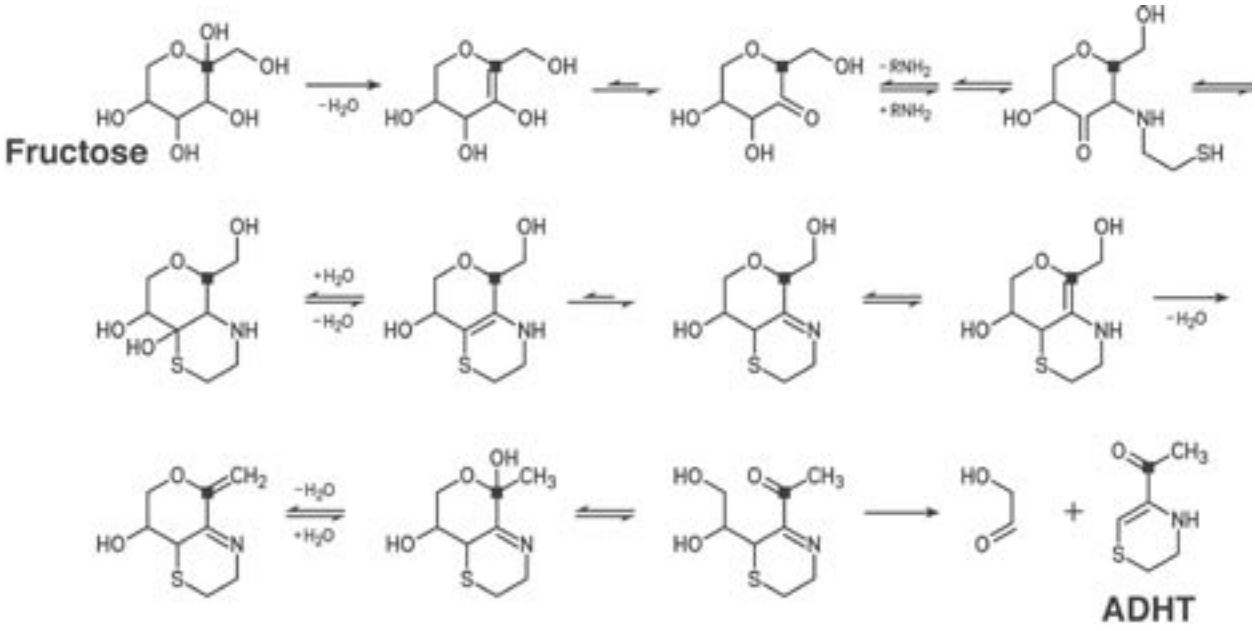


Figure 1. Formation of ADHT under dry heating conditions

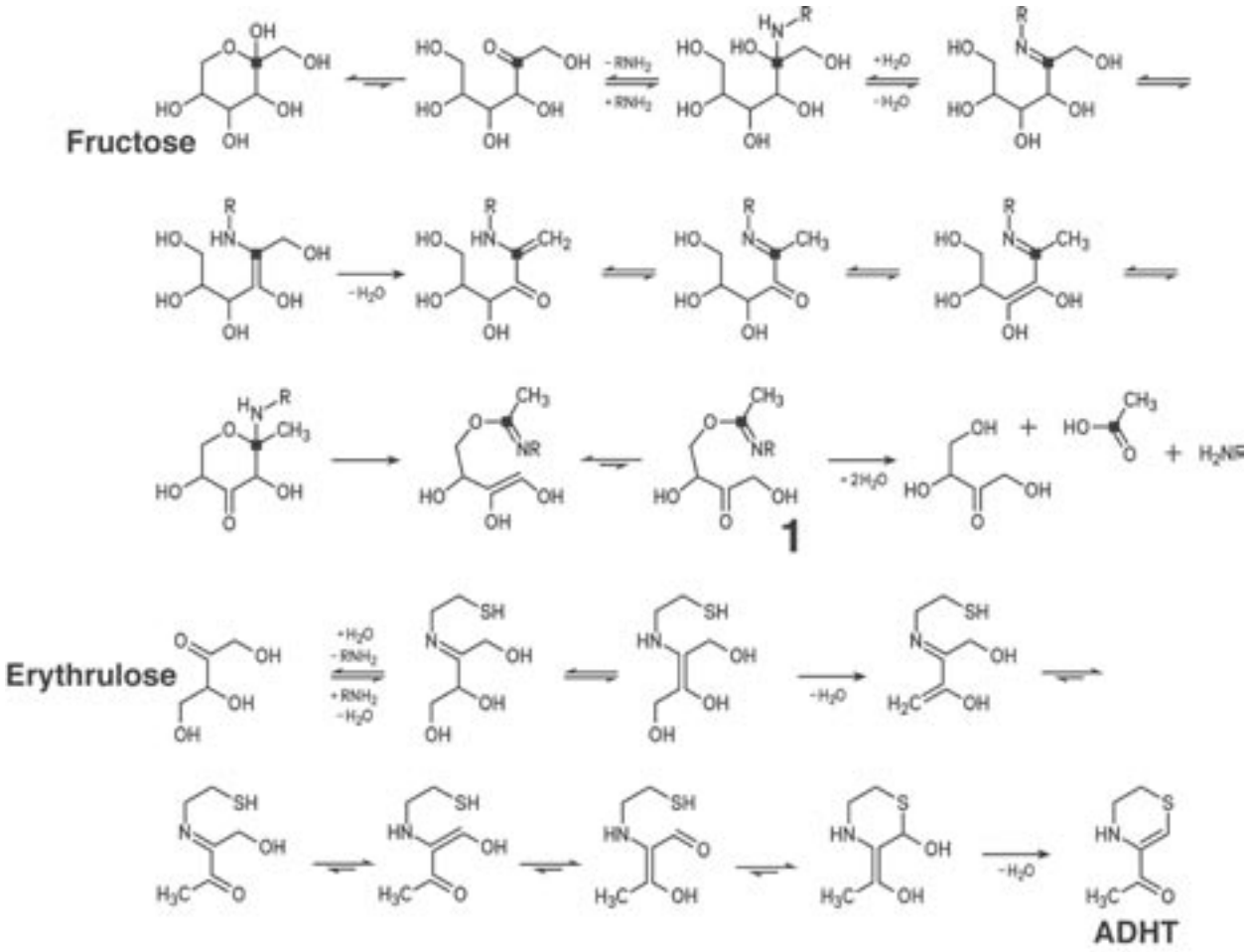


Figure 2. Formation of acetic acid and ADHT under cooking conditions

depicted in Figure 1 can alternatively also take place starting with the furanose form of fructose.

Contrary, under cooking conditions the well known formation of the Amadori product takes place. In the following steps, after ring closure 1 (Figure 2) is formed from which elimination of acetic acid is very reasonable, avoiding the awkward mechanisms postulated usually.

References

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