



Influence of pH, autolysis time and temperature on meaty volatile compounds formation of yeast autolysate

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Abstract

Meaty flavors produced from non-animal source is a market demand to the growth of the food products. The objectives of this research were effect of pH, autolysis time and temperature on meaty aroma compounds formation of yeast autolysate (YA). Heating suspension of dried yeast cell at different conditions of 50°C and 60°C at pH 3.0, 4.0 and 5.0. The optimum temperature and pH of autolysis was 50°C at pH 3.0 which gave the highest autolysate yield (52.00±0.11% w/w). Protein contents of autolysate during 16-26 hours were measured by Lowry's method. The highest protein content was 1.34 mg/mL at 50°C for 24hr at pH 3.0. Aroma volatile compounds was characterized through headspace-solid phase micro-extraction gas chromatography-time of flight mass spectrometry (HS-SPME-GC-ToFMS). Meaty volatile compounds are generated through Maillard reaction in three stages of yeast autolysate production. 2,5-dimethyl pyrazine (roasted beef), dimethyl trisulfide (sulfurous, meaty), furfural (bread) were the major volatile compounds related to cooked meat flavor. The best condition for dried yeast autolysate (180°C) produced sulfur and nitrogen containing compounds such as dimethyl trisulfide. The information about volatile characterization of YA could benefit the food and flavor industry involved in the manufacturing of meaty flavorings for non-animal origin products.

Keywords: Yeast autolysate, Autolysis, Thermal treatment, Meaty volatile compounds, GC-ToFMS

Introduction

Yeasts are one of the most important microorganisms widely used in the food industry.

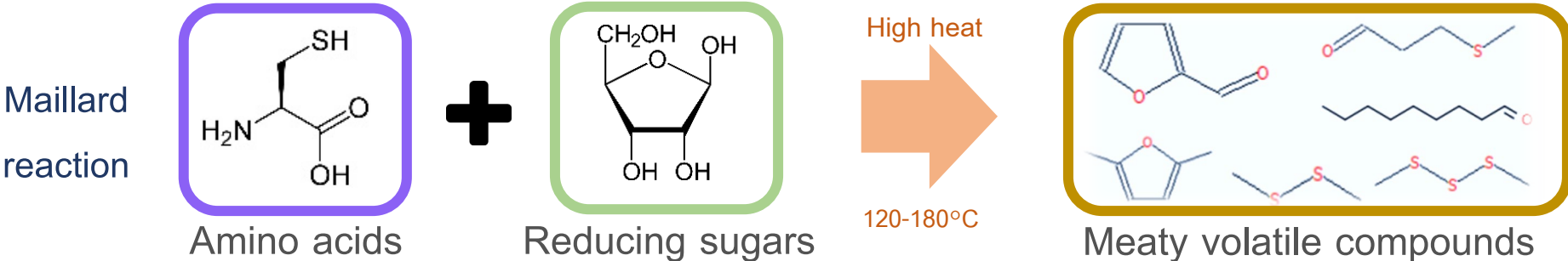
The value-added products from yeast are also widely used all over the world. Yeast extracts and autolyzed yeast extracts are categorically declared as GRAS (generally recognized as safe) by the Food and Drug Administration (FDA).

The rise in demand for meat flavoring agents has been attributed to the growth of the food industry.

Izzo and Ho (1991) investigated flavor compounds of yeast extract without adding beef protein hydrolysate and animal lipids. Heating is used to prepare the autolyzed yeast extract. It is environmentally friendly use of resources to lower the cost of meat flavoring production and encourages the development of meat flavoring agent from yeast.

Yeast extract (YE) is a natural flavoring agent obtained by extracting the liquid in yeast cells from *Saccharomyces cerevisiae* by hydrolysis. Hough and Maddox (1970) described yeast autolysate (YA) as a form of yeast extract obtained by autolysis of yeast cells.

Typically, the Maillard reaction products result from a complex reaction involving amino acids and reducing sugars, resulting in a significant alteration in the overall flavor profile of food systems. This reaction is widely regarded as the primary cause of many heat-treated food products' characteristic odorants. During the manufacturing of yeast autolysate (enzymatic hydrolysis, concentration, spray-drying, etc.), Maillard reaction occurs and impacts the odor and taste of yeast autolysate (Alim et al., 2018).



Until now, to our knowledge, little information is available concerning meaty volatile compounds generated in yeast autolysis production steps was reported. The objective of this research was to study the effect of autolysis pH, time, and temperature on meaty volatile compounds formation of YA during the production stages. This result will provide useful information for the quality control of the processing of YA liquid and powder related to meaty aroma compounds.

Table 2 Selected volatile compounds with relate to meaty flavor during production stages of yeast autolysate (YA) analyzed through HS-SPME-GC-ToFMS.

Aroma compounds	M.W. ^a	Identifi- cation	C.F. ^b	CAS no.	RI WAX ^c	Odor description ^d	Relative concentration ^e (µg/Kg)		
							Non-thermally treated YA ^f	Thermally treated YA ^f	Dried YA ^f
2-methylbutanal	86.1323	RI, MS	C ₅ H ₁₀ O	96-17-3	904	Cocoa	0.03 ± 0.02	2.32 ± 2.37	38.38 ± 11.36
3-methylbutanal	86.1323	RI, MS	C ₅ H ₁₀ O	590-66-3	909	Chocolate	0.02 ± 0.00	17.93 ± 6.46	209.04 ± 68.76
Dimethyl disulfide	94.20	RI, MS	C ₂ H ₆ S ₂	624-62-0	1063	Onion, cabbage, putrid	0.00 ± 0.00	0.01 ± 0.01	0.52 ± 0.44
Dimethyl trisulfide	126.264	RI, MS	C ₂ H ₆ S ₃	3658-80-8	1355	Sulfurous, meaty	0.00 ± 0.00	0.70 ± 0.41	0.21 ± 0.20
2,5-Dimethyl-Furan	96.1271	RI, MS	C ₇ H ₁₀ O	625-96-5	1288	Meaty, roasted beef	0.00 ± 0.00	0.00 ± 0.00	0.05 ± 0.00
Trimethyl-pyrazine	122.1677	RI, MS	C ₇ H ₁₀ N ₂	14667-55-1	1388	Roasted, potato, must	0.07 ± 0.06	0.03 ± 0.02	0.00 ± 0.00
2,5-Dimethyl-Pyrazine	108.1411	RI, MS	C ₆ H ₈ N ₂	123-32-0	1308	Roasted beef	0.03 ± 0.03	0.00 ± 0.00	0.11 ± 0.10
2,6-Dimethyl-Pyrazine	108.1411	RI, MS	C ₆ H ₈ N ₂	108-59-9	1314	Roasted nut	0.00 ± 0.00	0.00 ± 0.00	0.06 ± 0.00
2-Ethyl-3-methyl-Pyrazine	122.1677	RI, MS	C ₈ H ₁₀ N ₂	15707-23-0	1383	Roasted	0.00 ± 0.00	0.00 ± 0.00	0.03 ± 0.00
Furfural	96.084	RI, MS	C ₅ H ₄ O ₂	98-01-1	1452	Bread, almond, sweet	0.64 ± 0.17	308.87 ± 292.15	92.81 ± 2.47

^a Represents the molecular weight of aroma compounds.

^b Represents the chemical formula of aroma compounds.

^c Retention index of volatile compounds were calculated with n-alkanes mixture on a polar StabilWax[®] column.

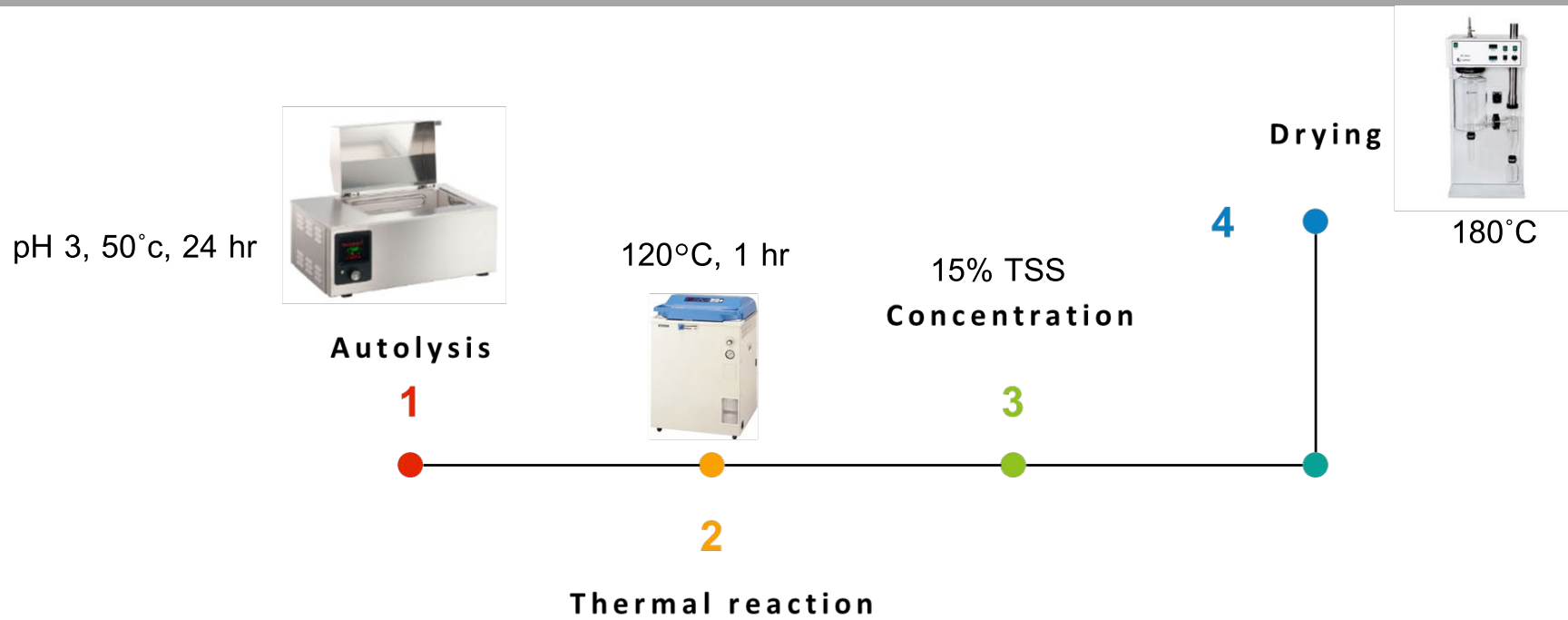
^d The odor property of a volatile compound was confirmed through online data bases; <http://flavornet.org>, www.thegoodscentscompany.com.

^e Relative concentration was calculated from concentration of internal standard (2-methyl-3-heptanone).

^f Non-thermally treated YA = YA at 25°C without thermal treatment; thermally treated YA = YA with thermal treatment at 120°C for 1 hour; dried YA = YA after thermal treatment at 120°C for 1 hour and then spray drying at inlet air temperature of 180°C.

All experiments performed in the table were conducted with three replications.

Materials and Methods



Results and Discussions

Autolysate yield

Table 1 Autolysate yield after autolysis at temperature of 50°C and 60°C at pH 3.0, 4.0 and 5.0 for 24 hr.

Temperature	pH	Autolysate yield % (w/w)
50°C	3.0	52.00±0.11
	4.0	36.00±0.23
	5.0	49.57±0.92
60°C	3.0	25.30±0.37
	4.0	17.63±0.92
	5.0	23.54±0.28

YA under 50°C at autolysis pH 3.0, 4.0, and 5.0 could give autolysate yield in the range of 36-52% w/w, which higher than 60°C. While yeast was autolyzed at pH 3.0, 50°C could give the highest autolysate yield (52.00±0.11% w/w).

Protein content during autolysis

The influence of pH and autolysis duration on protein content is given in Fig 1. Protein content was affected by autolysis pH and time. The concentration of protein released into liquid yeast autolysate from cells during autolysis at pH 3 considerably increased by 24 hr of incubation when the process was performed at temperature of 50°C. After 24 hr there was no appreciable increase at pH 5 of yeast autolysate. Therefore, 24 hr was used as autolysis time in this study. The highest protein contents were obtained as 1.34 mg/mL at pH 3 for 24 hr of autolysis and as 1.12 mg/mL at pH 5 for 16 hr of autolysis.

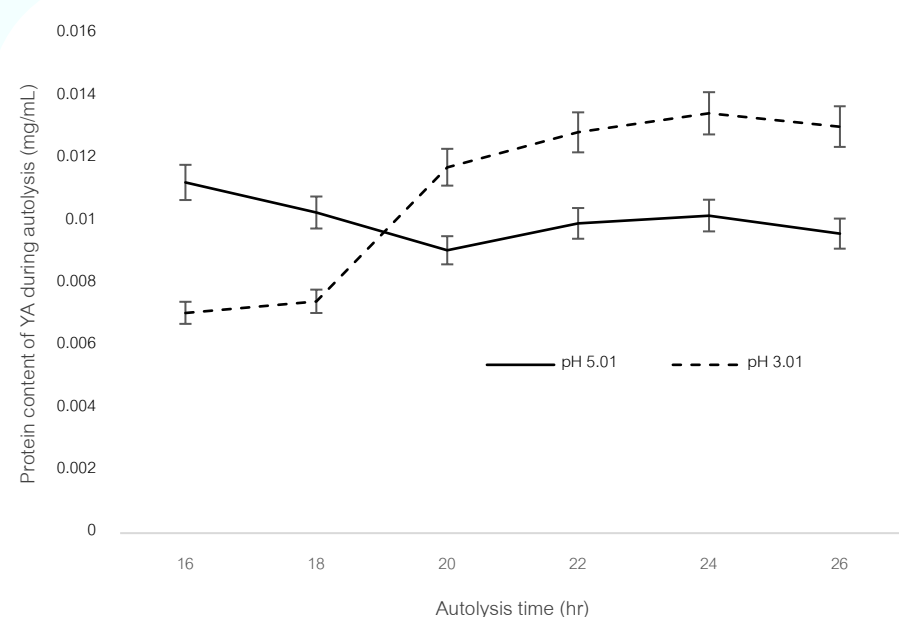


Fig 1 The influence of autolysis duration on protein content of yeast autolysate

Meaty volatile compounds

A total number of 5 meaty volatile compounds were identified in non-thermally treated YA sample (Table 2). It contained the highest relative concentration of volatile compounds which are acetic acid (sour), furfural (bread, almond, sweet) and butanoic acid (cheesy) respectively. It can be seen that YA sample before heating is exhibited little pyrazines and high amount of acetic acid (12.5 µg/L).

Table 2. show a total of 10 kinds of detected meaty volatile compounds from non-thermally treated, thermally treated and dried YA. The

following volatile compounds were identified in YA samples, including four pyrazines: trimethylpyrazine (roasted, potato, must), 2,5-dimethylpyrazine (roasted beef), 2,6-dimethylpyrazine (roasted nut), 2-ethyl-3-methylpyrazine (roasted); two furan derivative: 2,5-dimethylfuran (meaty, roasted beef), furfural (bread, almond, sweet); two aldehydes: 2-methylbutanal (cocoa), 3-methylbutanal (chocolate); two sulfur volatile compounds: dimethyl disulfide (onion, cabbage, putrid) and dimethyl trisulfide (sulfurous, meaty).

The present study has recorded 4 pyrazines in dried YA samples, these includes, 2,5-dimethylpyrazine, 2,6-dimethylpyrazine, 2-ethyl-3-methylpyrazine, and trimethylpyrazine. Pyrazines were generated in this experiment at thermal treatment temperature of 120°C and drying temperature of 180°C. Pyrazines is a chemical group which mostly generate after thermal reaction. With the increase in temperature, odor characteristics like meaty and roasted became more intense, when temperature reached 180°C, roasted, sweet and sulfurous odors were becoming predominant and stronger than other odors. As a result of the greater temperature of spray drying, various meaty aroma molecules are formed.

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Conclusion

Among the experiments, autolysis at 50°C for 24 hr at pH 3.0 was desirable condition in terms of autolysate yield and protein content. The highest levels of autolysate yield and protein content were 52.00±0.11% w/w and 1.34 mg/mL, respectively. Therefore, yeast autolysate was produced at those conditions for aroma compounds analysis. After the treatment of YA, it was observed that some volatile compounds such as dimethyl disulfide, dimethyl trisulfide, 2,5 - dimethylpyrazine, 2,6-dimethylpyrazine, trimethylpyrazine, 2-ethyl-3-methylpyrazine, 2-ethyl-5-methylpyrazine, furfural, 2,5- dimethylfuran were identified in YA samples in this study. Dried YA samples were shown the highest kinds and the concentration of volatile compounds. The GC-ToFMS analysis confirmed that the nitrogen and sulfur-containing volatile compounds were the main aroma compounds responsible for the meaty aroma of thermally treated products. The information about volatile characterization of YA could benefit the food and flavor industry involved in the manufacturing of meaty flavorings for non-animal origin products.

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