Available online at www.sciencedirect.com

ScienceDirect

Procedia Environmental Sciences 23 (2015) 400 – 406

Procedia Environmental Sciences

International Conference on Tropical and Coastal Region Eco-Development 2014(ICTCRED 2014)

Isolation and Identification Odorous Chemical Markers of Wastewater Poultry Slaughterhouse

Yoso Wiyarno*, Sri Widyastuti,

Department of Environmental Engineering, Faculty of Civil Engineering and Planning, University PGRI Adi Buana Surabaya, Indonesia

Abstract

Poultry slaughterhouse produces liquid waste from the blood of a chicken, the process of dyeing, washing chicken and production equipment in its production process. These wastes can act as a medium for microbial growth and development so that the waste is susceptible to decay. Besides causing a foul-smelling gas, the presence of dissolved oxygen excessive use can lead to a lack of oxygen for aquatic biota. Research has been done by capturing the smell of poultry slaughterhouse (*Rumah Pemotongan Ayam*/RPA) wastewater by using a modified smell extractor. Later, the smell was isolated through identification using GCMS (Gas Chromatography and Mass Spectrometry) to determine the chemical components as the composition of the sewage smell. It can be determined that the threshold value can be recommended as a threshold value that should not be exceeded at a chicken slaughterhouse wastewater or sewage treatment plant for waste is the dominant type of fat and protein. The components of successful compound identification using a library of GCMS is 1,2-Benzenedicarboxylic acid, 1,2-Bis(O-hydroksibenzo nitric)-cyclopropana, Phenantrene, Sulfur (molecule), 5-methoksi-2-metil-4-oxo-1,2,3,4-tetrahidro-1,10-phenantrolin, 4-chlorophenoxyacetic acid methyl ester, Phenol,2,2,-methylenbis(6-(1,1-dimethylethyl)-4-methyl-, Bis(2-ethylhexyl)phthalate, 4-methylthymidine, N-Cyano-N-N-N-N-tetramethyl-1,3,5-triazinetriamine, Diethyl bis(trimethylsilyl) ester

Peer-review under responsibility of scientific committee of the ICTCRED 2014

Keywords: liquid waste of chicken slaughterhouse, smell components, modification extractors

*Corresponding author.
E-mail: yoso.wiyarno@gmail.com and rafirudi@yahoo.co.id,

1. Introduction

Poultry slaughterhouses generate wastewater mainly from the slaughtering process, equipment and washing facilities as well as from production. This wastewater is characterized by large quantities of suspended solids, oil

Peer-review under responsibility of scientific committee of the ICTCRED 2014
doi:10.1016/j.proenv.2015.01.057

and grease, nitrogen and phosphorus which may vary depending on the process.[1] Proteins from carcass debris and blood are the major pollutants besides fat in poultry processing wastewater. With an estimated 2-5 % of total carcasses protein lost in the effluent, wastewater contains predominantly 35 % of protein, resulting in much higher biological oxygen demand (BOD) and chemical oxygen demand (COD) than town sewage.[2]

Poultry protein contains a total of 26 amino acids, of which only 2 namely L-Cysteine and L-Methionine are sulphur containing molecules and these constitute a minuscule percentage of the waste stream from a process plant. These are therefore not the principal sources of sulphur of hydrogen sulphide gas.[3] The basic causes of odour in poultry slaughterhouse wastewater stream are hydrogen sulphide gas, volatile organic sulphur compounds such as mercaptans, volatile fatty acids and nitrogenous compounds and organic particulate material. Methane and carbon dioxide, which are also almost always present, have no smell[4] of the principal odorous substances, hydrogen sulphide gas has the characteristic smell of rotten eggs and mercaptans smells like bottled LPG.

Based on the background described, the objective of the research is the isolation of volatile compounds into components making up the smell of a chicken slaughterhouse wastewater in the tub after a five-hour deposition by extraction modification, because the liquid waste after five hours of being on the vessel began a process of decay and smells.[5]

Identifying the components making up the smell of wastewater compounds chicken slaughterhouse is by investigating the deposition bath after five hours with Gas Chromatography (GC) and GCMS. Referring to the MOE Decree No. 50 of 1996 regarding the quality standards of the odor, mentioned several components of the compound with a concentration threshold which is considered as the source of the smell. However MOE Decree No. 50 of 1996 regarding the quality standards of the odor is a common rule for all the wastewater smell, and how the components of other compounds that are not contained in the rule, but is a major component in a waste constituent smells like the liquid waste home cuts of chicken) or a strong odor.[6] At what minimum concentration of compound components making up the smell of wastewater is still disturbing the public.

2. Research Method

2.1. Research Instrument:

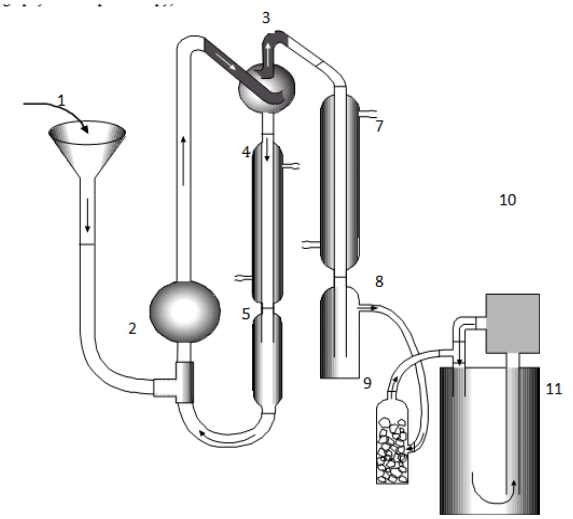
A set of extraction tools are modified as shown in the figure, the water suction pump that has been modified, laboratory glassware, Gas chromatography Hewlett Packard model 5890 Series with integrators and GCMS (Gas Chromatography Mass Spectroscopy).

2.2. Research Material:

The materials used in this study are taken from a chicken slaughterhouse wastewater in Surabaya. Other material used is Sodium sulphate anhydrous and Pentane.

2.3. Research Procedure:

The experiment is to determine the effectiveness of the extraction tool modifications to poultry slaughterhouse wastewater. The procedure of the experiment is as follows: first, poultry slaughterhouse wastewater with bottle samples is taken in the extract of the extractor tool as an insulator smell of wastewater. The extraction process is done for 4 hours. Liquid extract (still in the organic phase and the aqueous phase) is to separate the aqueous phase extracted from the organic phase using Pentane. Next, the organic phase is analyzed by GC (Gas Chromatography) and GCMS (Gas Chromatography Mass Spectroscopy) for the identification of the components making up the smell of wastewater compounds poultry slaughterhouse. Then, injection of as much as 1 mL into the GC-MS instrument performed, respectively to: (1) air (without sample and solvent), (2) solvent pentane. Finally, identification and confirmation of the identity of the volatile components is performed with a computer through a process of pattern matching mass spectra of each *atung* seed volatile components are separated by using a collection of mass spectra from NIST (National Institute of Standards and Technology) -USA.



Description:

1. The entrance of the effluent sample
2. Pumpkin for sample reservoir
3. Speed bowl
4. Tube for sample drainage equipped with cooler
5. Tube for holding sample
6. Connecting
7. Parallel Leibig cooler
8. Container for odor evaporation
9. Container for odor extraction
10. Vacuum (water pump)
11. Water reservoir

Figure 1. Extraction tool modification

3. Analysis and Discussion

3.1 Analysis of volatile components chicken slaughterhouse wastewater

GC chromatogram of a GC tool -MS 3 isolates injection results with Pentane solvent shows there are 11 components of the compound. The following compounds are associated with bad odors: mercaptans, skatoles, indoles, inorganic acids, aldehydes, ketones and organic compounds containing nitrogen or sulfuric atoms. These compounds can originate from the anaerobic decomposition of compounds with a high molecular weight, especially proteins. These are recognized as being among the causes of bad-smelling odors at the outlet of sewer lines and in treatment plants in general. Among the inorganic compounds, ammonia and hydrogen sulfide are considered to be the main causes of odor when the sewage comes from mainly households. The presence of hydrogen sulfide is caused by a reducing environment, i.e. characterized by low values of the oxidation-reduction potential.[7] This allows for the oxidation and reduction of methanthione and other proteins which readily breakdown to methyl mercaptan, dimethyl sulphide and H_2S.[8]

Proteins are biological macromolecules, either soluble or solid (e.g., feathers, hair, nails). It works outside the cell at an acid pH or in the presence of enzymes, soluble proteins precipitate, e.g., precipitation of casein by addition of rennet enzyme.

Table 1.
Result of the organic analysis making up the smell of wastewater compounds poultry slaughterhouse

Peak Number	Retention Time	Total Percentage	Compound Name
1	43,894	4.656	1,2-Benzenedicarboxylic acid
2	45,453	5.983	1,2-Bis(O-hidroksibenzo nitril)-cyclopropana
3	46,064	4.680	Phenantrene
4	46,752	5.115	Sulfur (molecule)
5	47,226	6.615	5-methoksi-2-metil-4-oxo-1,2,3,4-tetrahidro-1,10-phenantrolin
6	51,643	4.413	4-chlorophenoxyacetic acid methyl ester
7	53,775	33.433	Phenol,2,2,-methylenbis(6-(1,1-dimethylethyl)-4-methyl-
8	55,731	8.314	Bis(2-ethylhexyl)phthalate
9	58,131	7.231	4-methylthymidine
10	61,042	7.164	N-Cyano-N-N-N-N-tetramethyl-1,3,5-triazinetriamine
11	63,778	12.396	Diethyl bis(trimethylsilyl) ester

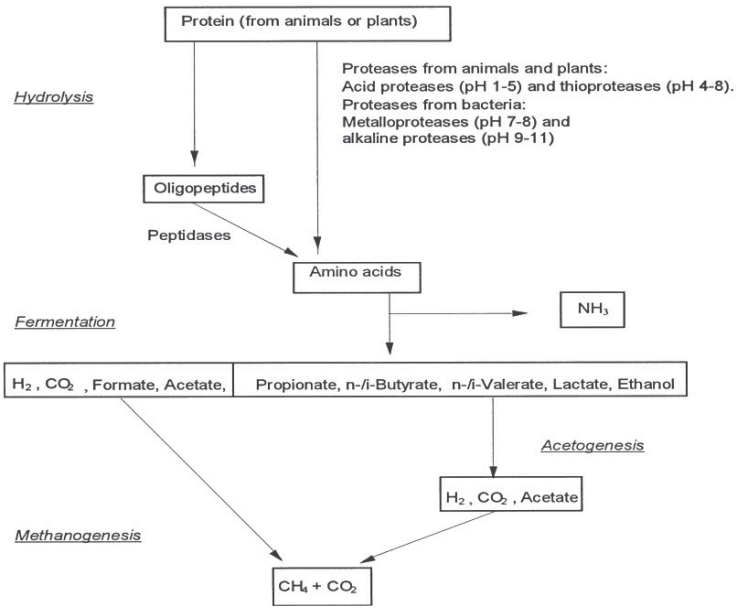


Figure. 2. Anaerobic degradation of proteins.[9]
(taken from Hans-Joachim Jördening, Josef Winter and Claudia Gallert, 2005)

In contrast to the fermentation of carbohydrates, which lowers the pH due to volatile fatty acid formation, fermentation of amino acids in wastewater reactors does not lead to a significant pH change, due to acid and ammonia formation. Acidification of protein-containing wastewater proceeds optimally at pH values of 7 or higher,[10] and ammonium ions together with the CO_2–bicarbonate–carbonate buffer system stabilize the pH. Acetogenesis of fatty acids from deamination of amino acids requires a low H_2 partial pressure for the same reasons as for carbohydrate degradation. This can be maintained by a syntrophic interaction of fermentative, protein-degrading bacteria and acetogenic and methanogenic or sulfate-reducing bacteria. Except for syntrophic interaction of amino acid-degrading bacteria with methanogens for maintenance. One amino acid, e.g., alanine (Eq. 4) is oxidatively decarboxylated and the hydrogen or reducing equivalents produced during

this reaction are used to reductively convert another amino acid, e.g., glycine, to acetate and ammonia (Eq. 5).

$$CH_3–CHNH_2–COOH + 2\ H_2O \rightarrow CH_3–COOH + CO_2 + NH_3 + 2\ H_2 \quad (1)$$

$$CH_2NH_2–COOH + 2\ H_2 \rightarrow 2\ CH_3COOH + 2\ NH_3 \quad (2)$$

$$\Delta G^{\circ}b = -38.9\ kJ\ mol^{-1}$$

For complete degradation of amino acids in an anaerobic system therefore, a syntrophism of amino acid-fermenting anaerobic bacteria with methanogens or sulfate reducers is required[11-13]. If long-chain amino acids are deaminated (Eqs. 6–9), fatty acids such as propionate, *i*-butyrate, or *i*-valerate are formed directly. The fatty acids require acetogenic bacteria for their degradation.

$$\text{valine} + 2\ H_2O \rightarrow \textit{i}\text{-butyrate} + CO_2 + NH_3 + 2\ H_2 \quad (3)$$

$$\text{leucine} + 2\ H_2O \rightarrow \textit{i}\text{-valerate} + CO_2 + NH_3 + 2\ H_2 \quad (4)$$

$$\textit{i}\text{-leucine} + 2\ H_2O \rightarrow \text{2-methylbutyrate} + CO_2 + NH_3 + 2\ H_2 \quad (5)$$

$$\text{glutamate} + 2\ H_2O \rightarrow \text{propionate} + 2\ CO_2 + NH_3 + 2\ H_2 \quad (6)$$

In contrast to carbohydrate degradation, where the necessity for propionate- and butyrate-degrading acetogenic bacteria can be circumvented by substrate limitation , during protein degradation these fatty acids are a product of deamination, and their formation cannot be avoided by maintaining a low H_2 partial pressure. In the methanogenic phase there is no difference in methanogenic activity whether carbohydrates or proteins are fermented, except that the methanogens in a reactor fed with protein need to be more tolerant to ammonia and higher pH.

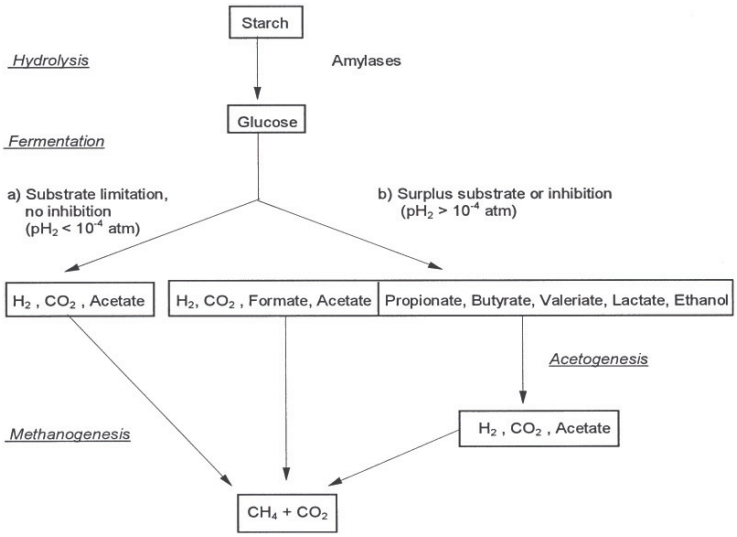


Figure. 3 Anaerobic degradation of starch under low- and high-loading conditions (taken from Hans-Joachim Jördening, Josef Winter and Claudia Gallert, 2005)

Fats and lipids are another group of biopolymers that contribute significantly to the COD in sewage sludge, cattle and swine manures, and wastewater from the food industry, e.g., slaughterhouses or potato chip

factories.[14-15] Glycerol and saturated and unsaturated fatty acids (palmitic acid, linolic acid, linolenic acid, stearic acid, etc.) are formed from neutral fats. Lipolysis of phospholipids generates fatty acids, glycerol, alcohols (serine, ethanolamine, choline, inositol), and phosphate. Lipolysis of sphingolipids generates fatty acids and amino alcohols (e.g., sphingosine), and li- polysis of glycolipids generates fatty acids, amino alcohols, and hexoses (glucose, galactose). A scheme for anaerobic degradation of fats is shown in Figure 3.
The long-chain fatty acids are degraded by acetogenic bacteria by ân oxidation to acetate and molecular hydrogen. If acetate and molecular hydrogen accumulate, the anaerobic digestion process is inhibited[16]. Odd numbered fatty acids are degraded to acetate, propionate, and hydrogen, and even numbered fatty acids to acetate and hydrogen[17]. Only at a very low H_2 partial pressure, which can be maintained by hydrogen utilizing methanogens or sulfate reducers, is â oxidation of at least *n*-butyrate or propionate exergonic. Methanol, ethanol, and ammonia are formed from choline (Fig. 4).

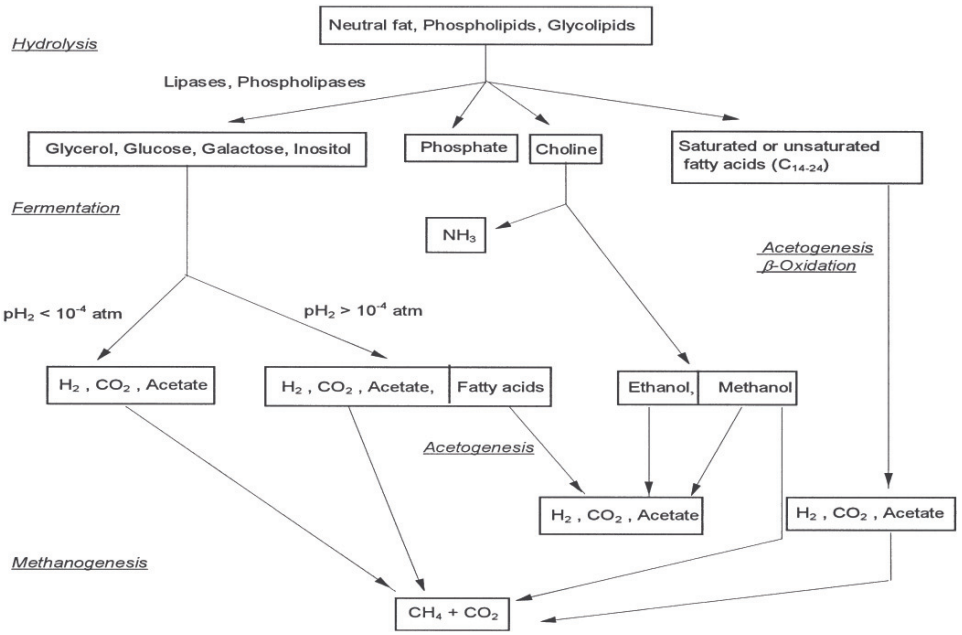


Figure 4 Anaerobic degradation of fats.
(taken from Hans-Joachim Jördening, Josef Winter and Claudia Gallert, 2005)

4. Conclusion

Eleven volatile components have been identified from three samples which are 1,2-Benzenedicarboxylic acid, 1,2-Bis(O-hidroksibenzo nitril)-cyclopropana, Phenantrene, Sulfur (molecule), 5-methoksi-2-metil-4-oxo-1,2,3,4-tetrahidro-1,10-phenantrolin, 4-chlorophenoxyacetic acid methyl ester, Phenol,2,2,-methylenbis(6-(1,1-dimethylethyl)-4-methyl-, Phenol,2,2,-methylenbis(6-(1,1-dimethylethyl)-4-methyl-, Bis(2-ethylhexyl)phthalate, 4-methylthymidine, N-Cyano-N-N-N-N-tetramethyl-1,3,5-triazinetria, Diethyl bis(trimethylsilyl) ester.

Acknowledgment

This research was funded by *Ditlitabmas*, the Directorate General of Higher Education, Ministry of National Education Research Grant Scheme of Fundamental Research for Fiscal Year 2014.

References

1. APTEC Technology Consulting (2009) Reducing Contamination & Odour in Poultry Slaughterhouses First of a Series of Articles on Causes & Cures for Odour, Contamination and Corrosion in the Poultry Industry Amaltas, Alstonia Estate, *Sigma IV*, Greater Noida 201308, India. http://aptec.in/wp-content/uploads/APTEC-Report-Size-Structure-of-Industry.pdf. Accessed on 29 June 2014
2. Del NV and Damianovic MHZ (2001) The Use of an Up-flow Anaerobic Sludge Blanket Reactor in the Treatment of Poultry Slaughterhouse Wastewater. Water Sci. Technol., 44: 83-88. http://www.iwaponline.com/wst/04404/wst044040083.htm Accessed on 29 June 2014
3. Hanaki, K., Matsuo, T., Nagase, M. (1981). Mechanism of inhibition caused by long-chain fatty acids in anaerobic digestion process. *Biotechnology and bioengineering*, **23**, 1591-1610. http://onlinelibrary.wiley.com/doi/10.1002/bit.260230717/abstract . Accessed on 2 July 2014
4. Hans-Joachim Jördening, Josef Winter and Claudia Gallert (2005) Environmental Biotechnology: Concepts and Applications Chapter 1. Bacterial Metabolism in Wastewater Treatment Systems Copyright © 2005 Wiley-VCH Verlag GmbH & Co. KGaA DOI: 10.1002/3527604286.ch1 http://onlinelibrary.wiley.com/doi/10.1002/3527604286.ch1/summary Accessed on 2 July 2014
5. Giancarlo Riva (2009) Sewage and Wastewater Odor Control, *Ozono Elettronica Internazionale*, Muggio, Italy Anthony Sacco, Spartan Environmental Technologies, Mentor, OH, USA **(www.spartanwatertreatment.co** Accessed on 2 July 2014
6. Guidelines on Odour Pollution and Its Control (2008) Central Pollution Control Board Ministry of Environment & Forests, Govt. of India Parivesh Bhawan, East Arjun Nagar, Delhi–1100 32 http://cpcb.nic.in/divisionsofheadoffice/pci2/package_odourreport_2.12.08.pdf Accessed on 2 July 2014
7. MOE Decree No. 50 of 1996 Regarding the Quality Standards of the Odor http://www.cets-uii.org/BML/Kebauan/kepmen5096/ Accessed on 2 July 2014
8. Odour and Environmental Engineering Consultants (2008) www.odourireland.com Accessed on 2 July 2014
9. Sheridan, BA, Hayes, E.T., Curran, T.P., Dodd, VA, (2004). A dispersion modelling approach to determining the odour impact of intensive pig production units in Ireland. *Bioresource Technology*. Published.
10. Yordanov, D. (2010) Preliminary Study of the Efficiency of Ultrafiltration Treatment of Poultry Slaughterhouse Wastewater *Bulgarian Journal of Agricultural Science,* 16 (No 6) 2010, 700-704
11. Agricultural Academy University of Food Technology, Department of Meat and Fish Technology, BG - 4002 Plovdiv, Bulgaria. *http://www.agrojournal.org/16/06-06-10.pdf.*. Accessed on 3 July 2014
12. ZhangZH, Barlow JN, Baldwin JE, Schofield CJ. (1997). Metal-catalyzed oxidation and mutagenesis studies on the iron(II) binding site of 1-aminocyclopropane-1-carboxylate oxidase. *Biochemistry 36,*15999–16007. http://www.chem.qmul.ac.uk/iubmb/enzyme/EC1/14/17/4.html Accessed on July 2014