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# Chemical and Biochemical Aspects of Slaughterhouse Sludge Intended for Feed Purposes

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The protein and fat qualities of both chemically flocculated and aerobically activated sludge from pig and poultry slaughterhouses were investigated. Protein and total volatile nitrogen levels varied considerably between and within sludge types and animal species slaughtered. The biogenic amines cadaverine, histamine, and putrescine could be detected at various levels. Iron chloride, used as a flocculant, resulted in high peroxide values (148–568 mequiv/kg of fat). Low peroxide values were detected in aerobically activated sludge and chemically flocculated sludge in which calcium lignosulfonate was used as a flocculant. Free fatty acid (FFA) concentration ranged from 5 to 10% in activated sludge and from 10 to 71% in chemically flocculated sludge. Binding of $Ca^{2+}$ from calcium lignosulfonate with FFA most probably accounted for the high level of FFA detected in this flocculated sludge.

**Keywords:** *Flocculants; slaughterhouse sludge; chemical composition; pig feed*

## INTRODUCTION

To improve logistics and to meet consumer demand for cheap meat, slaughter processes have been intensified and centralized at a limited number of locations. As a result, the amounts of water used at a single slaughterhouse have increased. Earlier investigations revealed that approximately 2–5% of the total carcass protein of slaughtered animals is lost in the process water (Grant, 1976). The costs for discharging untreated slaughterhouse process water into the local sewage systems have increased during the past decennia. The regulations for discharging process water into surface water have been tightened as well. Therefore, many slaughterhouses have decided to (pre)treat the process water themselves (Heusinkveld, 1989). In general, two types of water treatment methods are applied. There is the flocculation–flotation method, which is a physicochemical method in which flocculants are used, and a biological method in which aerobically activated sludge is used. As a result of flocculation–flotation, the organic part of the dry matter in chemically flocculated sludge from slaughterhouses mainly consists of protein and fat from animal origin (Mulder *et al.*, 1986). As a result of the biological water treatment, the organic part of the dry matter in aerobically activated sludge mainly consists of protein (single cell) and to a lesser degree fat (Kavanagh *et al.*, 1978). The chemical content of both chemically flocculated sludge and activated sludge resembles that of other feed constituents such as soybean meal or meat meal (Mulder *et al.*, 1986; NRC, 1984, 1988; Heddle, 1979; Kavanagh *et al.*, 1978).

Flocculated and activated sludges from slaughterhouses are highly contaminated with human and animal pathogenic bacteria, as well as flora that can spoil the product (Mulder *et al.*, 1986; Fransen *et al.*, 1995). Thus, deterioration of protein (Urlings *et al.*, 1993) and fat by proteolytic and lipolytic activities or oxidation of fats and fatty acids can be expected (Mulder *et al.*, 1986). Concerning chemically induced processes in chemically flocculated sludge, Black *et al.* (1992) performed an experiment in a laboratory model of a dissolved air flotation (DAF) unit with $Fe_2(SO_4)_3$ used as a flocculation aid (M. D. Black, Kemin Industries, Iowa, personal communication, 1993). These results (Black *et al.*, 1992) revealed that oxidation of fats predominantly occurred during flotation, resulting in an increase of the peroxide value from 33 ($t = 0$) to 190 mequiv/kg of fat ($t = 60$ min). Furthermore, in chemically flocculated poultry and pig sludge, high levels of zinc, copper, and iron were found (Mulder *et al.*, 1986). As binding of metals is enhanced in activated sludge flocs (Sterritt and Lester, 1981; Norberg and Persson, 1984; Rossin *et al.*, 1982), high concentrations of these metals are to be expected in activated slaughterhouse sludge as well.

To investigate the possibility of processing poultry and pig sludge into a wet feed constituent for pigs, the quality of both chemically flocculated and aerobically activated raw sludge from pig ($n = 8$) and poultry ($n = 5$) slaughterhouses was surveyed. Water treatment process parameters, such as type of water treatment process and duration, were considered. The formation of biogenic amines and ammonia as a result of protein and successive amino acid breakdown was determined. Concentrations of iron, zinc, copper, lead, and cadmium were determined as well. In addition, oxidation of fats and formation of free fatty acids by lipolytic activities were measured. Possible effects of process parameters on protein and fat quality are discussed.

## MATERIALS AND METHODS

**Slaughterhouses.** Five poultry slaughterhouses (companies A–E) and eight pig slaughterhouses (companies F–M) were involved in this survey. Per location the amount of animals slaughtered on a daily basis ranged from 48 000 to 120 000 broilers and from 3000 to 7000 pigs, respectively.

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**Table 1. Species Slaughtered at the Different Locations, Water Treatment Processes Applied, Mean Duration of Water Treatment Processes, Flocculants Used, and Current Sludge Destination**

| Co. | species | water treatment process | mean duration of water treatment process | flocculants | current sludge destination: flocculated sludge | current sludge destination: activated sludge |
|---|---|---|---|---|---|---|
| A | poultry | flocculation−flotation | 20−40 min | $FeCl_3$ + polym[a] | fertilizer | |
| B | poultry | flocculation−flotation | 20 min | $FeCl_3$ + polym | rendering plant | |
| C | poultry | flocculation−flotation | 20 min | $FeCl_3$ + polym | fertilizer | |
| D | poultry | flocculation−flotation/aerobically activated sludge | 6 + 18 h | $FeCl_3$ + polym | rendering plant | recycled in flotation unit |
| E | poultry | flocculation−flotation/aerobically activated sludge | 15 min + 1.5 days | $FeCl_3$ + polym | rendering plant | recycled in flotation unit |
| F | pigs | flocculation−flotation | 2 h | $FeCl_3$ + polym | rendering plant | |
| G | pigs | flocculation−flotation | 20 min | $FeCl_3$ + polym | fertilizer | |
| H | pigs | aerobically activated sludge/flocculation flotation | (14 h +) 30−45 min | $FeCl_3$ | fertilizer | unknown |
| I | pigs | flocculation flotation/aerobically activated sludge | 2.5−4 h + 7 days | CLS + SHMF + SA[b] | rendering plant | fertilizer |
| J | pigs | aerobically activated sludge | 6 days | | | fertilizer |
| K | pigs | aerobically activated sludge | 5 days | ($FeCl_3$)[c] | | fertilizer |
| L | pigs | aerobically activated sludge | 1 day | | | fertilizer |
| M | pigs | aerobically activated sludge | 1.5 days | polym | | fertilizer |

[a] Polymeric solution. [b] Calcium lignosulfonate + sodium hexametaphosphate and sulfuric acid. [c] $FeCl_3$ was used as a flocculation aid, because of problems concerning sedimentation.

**Water Treatment Facilities at the Slaughterhouses.** Table 1 presents the water treatment processes performed, mean water treatment duration, and flocculants used at the different slaughterhouses. Sludge originating from the aerobically activated water treatment process from companies D and E was recycled in the flocculation−flotation process. At company I both chemically flocculated ($I_1$) and aerobically activated ($I_2$) sludges were disposed of separately and were sampled accordingly. At slaughterhouses J and M process water was treated with an aerobically activated water treatment process, but purified water and sludge were separated by flotation instead of sedimentation. At slaughterhouses A−H iron chloride ($FeCl_3$, 40% solution) and different polymeric solutions were used as coagulants/flocculants during water treatment. At slaughterhouse I calcium lignosulfonate, combined with sodium hexametaphosphate and sulfuric acid ($H_2SO_4$), was used as flocculant. In company M a polymeric solution was used to prevent sedimenting of sludge during forced aeration.

**Sludge Samples.** Fresh chemically flocculated sludge samples (A−$I_1$) were collected directly after the sludge was mechanically separated from the water. Fresh aerobically activated sludge samples ($I_2$−M) were collected after primary sedimentation. Samples for the determination of free fatty acids and peroxide value were vacuum packed, cooled, and stored at 2−4 °C immediately on the spot. All samples were transported to the laboratory and kept at −20 °C pending chemical analyses. Analysis of the peroxide value was performed within 1 week after sampling. Samples were analyzed in duplicate.

**Chemical Analyses.** Dry matter and ash content was analyzed following the standard procedures (NSI, 1973a, 1978). Total nitrogen was analyzed by Kjeldahl (NSI, 1975) and represented as protein (N × 6.25). Total volatile nitrogen (TVN) was analyzed after precipitation with 10% trichloroacetic acid (TCA) according to Kjeldahl (Lindgren and Pleje, 1983). The concentration of the biogenic amines histamine, putrescine, and cadaverine in sludge was analyzed by HPLC using a Zorbax 300 SCX column (25 × 4.6 mm) and 0.1% phosphate buffer (pH 6.1)/methanol (70:30%) as the mobile phase. Detection was performed by fluorometry after postcolumn derivatization with *o*-phthalaldehyde. The samples were extracted by a 5% TCA solution as described by Kerstholt *et al.* (1983). The detection level was 1 mg/kg of sludge. Metal concentrations were measured in 25 mg samples after drying (105 °C for 7 days) and destruction of the sludge samples with 1 mL of 70% nitric acid in a water bath at 70 °C for 16 h. Iron, copper, and zinc were determined using the flame technique, and cadmium and lead were determined using the graphite oven technique. Standard procedures in a Perkin-Elmer Zeeman/5100 PC atomic absorption spectrometer were used for both flame and oven techniques (Kessels and Wensing, 1993). Free fat was determined according to a standard procedure (NSI, 1973b). For determination of free fatty acids and the peroxide value, fats were extracted with petroleum ether (30−60 °C)/methanol (2:1). The extract was filtered through a glass fiber filter pad (Baxter grade 391 or equivalent) and vacuum distilled at 30−40 °C for 15 min (Black, personal communication, 1993). To prevent oxidation during extraction, 0.05% 2,6-di-*tert*-butyl-4-methylphenol (BHT) was added to the petroleum ether/methanol mixture and $N_2$ gas was used to exclude oxygen. Free fatty acids and peroxide value were determined according to standard procedures (NSI, 1980a,b). Free fatty acids were measured as a percentage of free fat.

## RESULTS

The duration of the water treatment processes ranged from 20 min to 6 h for flocculation−flotation and from 14 to 168 h for the activated water treatment process (Table 1). $FeCl_3$ was used as a flocculant during the flocculation−flotation water treatment process in all but one slaughterhouse. In the latter, calcium lignosulfonate and sodium hexametaphosphate combined with sulfuric acid were used for flocculation.

Dry matter contents (DM), ash, protein, and percentage of total volatile nitrogen (TVN)/total nitrogen (TN) in chemically flocculated and activated sludge from pig and poultry slaughterhouses are summarized in Table 2. With the exception of company E, the dry matter content in chemically flocculated sludge ranged from 4.3 to 12.2%, whereas in activated sludge it ranged from 0.9 to 5.8%. The chemically flocculated sludge of company E was dehydrated by a bandpress and reached

**Table 2. Dry Matter (DM), Ash, and Protein Contents, Percentage of Total Volatile Nitrogen/Total Nitrogen, and Biogenic Amines (Cadaverine, Histamine, and Putrescine) in Chemically Flocculated (floc) and Aerobically Activated (act.) Sludge from Poultry and Pig Slaughterhouses (*n* = 1, Analyzed in Duplicate)**

| Co. | water treatment process | species | DM (%) | ash (g/kg of DM) | protein (g/kg of DM) | TVN/TN (%) | biogenic amines (mg/100 g of protein) | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | cadaverine | histamine | putrescine |
| A | floc | poultry | 7.2 | 110 | 417 | 1.4 | 189 | 75 | 48 |
| B | floc | poultry | 7.7 | 152 | 533 | 2.5 | 313 | 81 | 75 |
| C | floc | poultry | 8.9 | 91 | 471 | 3.1 | 167 | 73 | 83 |
| D | floc | poultry | 7.0 | 139 | 414 | 2.5 | 32 | 24 | 24 |
| E | floc | poultry | 18.0 | 195 | 625 | 3.1 | 19 | 11 | 11 |
| F | floc | pigs | 4.3 | 147 | 535 | 1.4 | 131 | 47 | 75 |
| G | floc | pigs | 12.2 | 114 | 369 | 7.6 | 28 | 82 | 82 |
| H | floc | pigs | 11.4 | 140 | 614 | 8.7 | 6 | nd* | 6 |
| $I_1$ | floc | pigs | 11.2 | 107 | 473 | 3.2 | 109 | 43 | 83 |
| $I_2$ | act. | pigs | 2.1 | 252 | 429 | 1.3 | nd | nd | 78 |
| J | act. | pigs | 2.2 | 200 | 504 | 1.9 | 218 | 60 | 79 |
| K | act. | pigs | 5.8 | 167 | 518 | 4.4 | 285 | 135 | 87 |
| L | act. | pigs | 3.1 | 184 | 481 | 6.3 | 132 | 35 | 69 |
| M | act. | pigs | 0.9 | 250 | 478 | 1.4 | 23 | 70 | 70 |

[a] nd, not detectable (<1 mg/kg of sludge).

**Table 3. Concentration of Iron (Fe), Copper (Cu), Zinc (Zn), Lead (Pb), and Cadmium (Cd) in Chemically Flocculated and Aerobically Activated Sludge from Poultry and Pig Slaughterhouses (*n* = 1, Analyzed in Duplicate)**

| Co. | water treatment process | concentration of trace elements (mg/kg of DM) | | | | |
|---|---|---|---|---|---|---|
| | | Fe | Cu | Zn | Pb | Cd |
| A | floc | 39 582 | 118 | 207 | 4.2 | 0.3 |
| B | floc | 41 389 | 86 | 199 | 4.1 | 0.2 |
| C | floc | 21 852 | 38 | 165 | 2.9 | 0.2 |
| D | floc | 41 036 | 167 | 550 | 84.3 | 0.4 |
| E | floc | 49 619 | 183 | 349 | 34.0 | 0.4 |
| F | floc | 35 509 | 91 | 286 | 5.0 | 0.3 |
| G | floc | 36 055 | 76 | 219 | 5.6 | 0.4 |
| H | floc | 49 444 | 192 | 449 | 45.8 | 0.3 |
| $I_1$ | floc | 3 617 | 139 | 136 | 3.6 | 0.2 |
| $I_2$ | act. | 7 953 | 406 | 481 | 10.4 | 0.5 |
| J | act. | 5 892 | 593 | 1105 | 53.4 | 1.2 |
| K | act. | 12 485 | 226 | 786 | 24.5 | 0.7 |
| L | act. | 10 554 | 306 | 978 | 41.1 | 1.3 |
| M | act. | 21 640 | 270 | 843 | 28.0 | 1.2 |

**Table 4. Fat Content, Peroxide Value, and Percentage of Free Fatty Acids in Flocculated and Aerobically Activated Sludge from Poultry and Pig Slaughterhouses (*n* = 1, Analyzed in Duplicate)**

| Co. | water treatment process | species | fat content (g/kg of DM) | peroxide value (mequiv/kg of fat) | free fatty acids (%) |
|---|---|---|---|---|---|
| A | floc | poultry | 354 | 170 | 24 |
| B | floc | poultry | 217 | 148 | 10 |
| C | floc | poultry | 124 | 568 | 59 |
| D | floc | poultry | 276 | 148 | 10 |
| E | floc | poultry | 250 | 203 | 43 |
| F | floc | pigs | 88 | 216 | 19 |
| G | floc | pigs | 292 | 355 | 34 |
| H | floc | pigs | 77 | 278 | 24 |
| $I_1$ | floc | pigs | 227 | 4 | 71 |
| $I_2$ | act. | pigs | 44 | 3 | 5 |
| J | act. | pigs | 14 | 32 | 5 |
| K | act. | pigs | 42 | 5 | 10 |
| L | act. | pigs | 33 | 12 | 10 |
| M | act. | pigs | 25 | 14 | 8 |

a DM content of 18%. The mean ash content in chemically flocculated sludge was lower than in activated sludge, 133 ± 31 and 195 ± 52 mg/kg of DM, respectively. The protein content of the sludge ranged from 37 to 63% of the dry matter. The TVN/TN% fluctuated considerably between and within sludge types and animal species slaughtered. In sludge of nearly all slaughterhouses biogenic amines could be detected, but at variable levels. In general, the concentration of cadaverine was higher than the concentration of histamine and putrescine.

Concentrations of trace elements in sludge are presented in Table 3. Iron concentrations were higher in chemically flocculated sludge (21−49 g/kg of DM) than in activated sludge (6−22 g/kg of DM), except for flocculated sludge from company I, in which the concentration of iron was relatively low (4 g/kg of DM). In chemically flocculated sludge concentrations of copper and zinc were generally lower compared to those in activated sludge. Large variations in lead concentrations were observed in sludge from different companies applying flocculation−flotation. Cadmium concentration in chemically flocculated sludge was lower than in activated sludge, 0.2−0.4 and 0.5−1.3 mg/kg of DM, respectively.

In Table 4 some parameters of the fat quality of the sludges are presented. The fat content in chemically flocculated sludge was higher than in activated sludge, 77−354 and 14−44 mg/kg of DM, respectively. In general, the peroxide value in chemically flocculated sludge was high, 148−568 mequiv/kg of fat, except for company $I_1$, in which the peroxide value in sludge was 4 mequiv/kg of fat. In activated sludge the peroxide value ranged from 3 to 32 mequiv/kg of fat. In general, free fatty acid concentration was about 10 times higher in chemically flocculated sludge compared to activated sludge, 10−71 and 5−10%, respectively.

## DISCUSSION

In contrast to earlier investigations (Mulder *et al.*, 1986), no significant differences in protein content between pig and poultry chemically flocculated sludges could be detected. The mean fat concentration found in flocculated poultry sludge (A−E) was comparable to the level found by El Boushy *et al.* (1984), 244 and 286 g/kg of DM, respectively. As was stated before (Mulder *et al.*, 1986), fat and protein together accounted for 60−88% of the dry matter in chemically flocculated sludge and for 47−57% in aerobically activated sludge. Besides the use of flocculants, the two main parameters that are different between the flocculation–flotation process and the activated sludge process, which can adversely affect protein and fat quality, are the process duration and the presence of a balanced microflora in the activated sludge. Both lead to protein and fat breakdown by microbial enzymatic activity during the aerobically activated water treatment process and probably account for the lower level of fat in activated sludge compared to chemically flocculated sludge. Compared to the flocculated sludge from companies G and $I_1$, the fat content of the flocculated sludge from slaughterhouse F and H was low. In the case of slaughterhouse H this was probably caused by lipolytic activity during the preceding aerobically activated water treatment. In the case of slaughterhouse F probably other, unknown, factors contributed to this observation. The phenomenon that the ash content in activated sludge was higher compared to that in chemically flocculated sludge (average 195 ± 52 g/kg of DM for activated sludge and 133 ± 31 g/kg of DM for chemically flocculated sludge) is probably a result of the breakdown of protein and fat during the biological water treatment, which results in relatively more ash residing per kilogram of dry matter.

Concerning the protein quality in sludge, the formation of biogenic amines and volatile compounds such as ammonia is important. Biogenic amines are formed predominantly as a result of the activity of amino acid decarboxylase(s) produced by bacteria (de Boer, 1988). Decarboxylases catalyze the conversion of amino acids into the corresponding amines, e.g. lysine into cadaverine, histidine into histamine, and arginine via ornithine into putrescine. Clostridia and Enterobacteriaceae were found to produce enzymes such as lysine decarboxylase, histidine decarboxylase, and ornithine decarboxylase (Beutling *et al.*, 1984; Behling and Taylor, 1982; de Boer, 1988). As these microorganisms were also present in sludge in considerable amounts (Fransen *et al.*, 1995; Mulder *et al.*, 1986), the formation of biogenic amines in sludge was to be expected. Probably, differences in water collecting and the drainage systems at the slaughterhouses, i.e. the time between water collection and water treatment, accounted for the different levels of biogenic amines in the sludge from the different slaughterhouses.

The level of histamine in sludge ranged from <0.1 (detection level) to 4.1 mg/100 g of sludge, thus being below the safety level of 5 mg % set by the FDA (Bartholomew *et al.*, 1987). Concerning the toxicity of cadaverine and putrescine, hardly any data are available. Nevertheless, potentiation of the toxicity of histamine by cadaverine and putrescine is mentioned in the literature (Bjeldanes *et al.*, 1978; Taylor, 1986). Although the level of histamine in sludge was below the (acute) toxicity level as set by the FDA, daily consumption of sludge as a feed constituent should be regarded as semichronic exposure. For that reason and also to assure the highest possible quality, microbial decarboxylation of amino acids into biogenic amines, especially essential amino acids such as lysine, should be prevented as much as possible.

In some sludge types the amino acid breakdown through deamination was considerable, TVN/total N ranging from 1 to 9%. For instance, Enterobacteriaceae or clostridia spp. are able to convert amino acids into ammonia (Meister, 1965; Barker, 1965). As these bacteria were present in raw sludge (Mulder *et al.*, 1986; Fransen *et al.*, 1995), they are likely to be responsible for the amino acid breakdown in sludge. High levels of ammonia can be toxic for mammals. A study performed with rats continuously intragastrically infused with an ammonia acetate solution revealed that a total dose up to 1 g of ammonia/kg of body weight/day had no lethal effects (Ingle and Williams-Ashman, 1962). With an average of 500 g of protein/kg of dry matter of sludge and a maximum TVN/total N level of 10%, the concentration of ammonia in sludge is approximately 0.05 g of $NH_4^+$/kg of sludge. Assuming that sludge would account for approximately 15–20% of a feed ration (on a dry matter base) of, for instance, pigs, then, depending on the dry matter content of the sludge, about 1–5 kg of sludge/day will be fed. Therefore, concerning ammonia, sludge can be regarded as safe. Nevertheless, when sludge is intended to be used as a valuable feed constituent, the protein quality should be preserved as much as possible, especially when essential amino acids are concerned. For that reason excessive microbial protein and amino acid breakdown should be avoided. This means that, for instance, the time between water collection, water treatment, and handling of the sludge should be minimized.

When $FeCl_3$ was used as a flocculant, iron and zinc levels were increased in flocculated sludge: 38 g of Fe/kg of DM and 0.3 g of Zn/kg of DM. This was comparable to the iron and zinc levels detected by El Boushy *et al.* (1984): 41 g of Fe/kg of DM and 0.4 g of Zn/kg of DM. In general, during the activated sludge process no flocculants are used, because the balanced system facilitates the separate processes of aeration and sedimentation occurring without problems. Nevertheless, in company K a certain amount of $FeCl_3$ was used, because of problems during sedimentation of sludge flocks, explaining the relatively high concentration of iron in the sludge. High concentrations of iron, zinc, lead, and cadmium in activated sludge from company M can be explained as a result of the use of soil water out of their own well. Metals were filtered, and the filters were washed three times a week, draining this wash water into the activated sludge plant. High concentrations of zinc, lead, and cadmium in activated sludge are a result of the binding of metals onto sludge flocks (Norberg and Persson, 1984; Oliver and Cosgrove, 1974; Rossin *et al.*, 1982, Sterrit and Lester, 1981). Although high concentrations of lead in sludge were suggested to be caused by polymeric solutions such as polyacrylamide, used as flocculation/sedimentation aids in different slaughterhouses (companies A–F and M), this could not explain the increased levels of lead in sludge originating from slaughterhouses H and J–L. The water from these slaughterhouses was treated with the aerobically activated water treatment process. It was suggested that other factors account for a high level of lead in sludge as well, e.g. plumbiferous paints, pipes, and seals present in these slaughterhouses.

The peroxide value reached high values during forced flotation, as has been seen before (Black *et al.*, 1992). High concentrations of metals, especially iron, and forced flotation by aeration will catalyze oxidation of fats, as was seen in the sludge types in which $FeCl_3$ was used as a flocculant. Activated sludge is firmly aerated as well, to provide the oxygen necessary for the conversion of the organic material present in the process water into bacterial cell material. The concentrations of copper, zinc, and lead in the activated sludge were relatively high, but the iron concentration was low. This resulted in peroxide values that were relatively low. Therefore, it was suggested that in aerobically activated sludge the relatively low iron concentration is the important factor accounting for the low level of fat oxidation. Breakdown of peroxides through peroxidases during the water treatment process, resulting in smaller components which cannot be detected with the applied analytical method (Belitz and Grosch, 1986), may be another explanation.

According to Mulder *et al.* (1986), a high degree of free fatty acids could be detected in chemically flocculated poultry and pig slaughterhouse sludge. The presence of free fatty acids in feedstuff is not unfavorable in view of the digestibility of fat. However, oxidation of free fatty acids is thought to occur more easily than oxidation of fatty acids bound in glycerides. This will result in rancidity of the fat. Compared to activated sludge, the concentration of free fatty acids was higher in chemically flocculated sludge. In flocculated sludge originating from slaughterhouse I ($I_2$) the concentration of free fatty acids was the highest detected, whereas the peroxide value remained low in the same sludge. It is suggested that $Ca^{2+}$ from calcium lignosulfonate binds free fatty acids into complexes that precipitate (Belitz and Grosch, 1986; El Boushy and van der Poel, 1994) and make them less accessible for oxidation. As lignosulfonates are biodegradable and are allowed without limit in feedstuffs as a binding agent (E565) (DPBF, 1992), calcium lignosulfonate seems to be an interesting substitute for $FeCl_3$.

Considering the average chemical composition of the sludge, i.e. protein and fat quality, it can be stated that chemically flocculated and aerobically activated sludges from poultry and pig slaughterhouses are potential feed constituents for pigs. The production of sludge under the recent circumstances can create some problems concerning the concentrations of elements such as iron, copper, and lead in sludge when it is used in the same order of magnitude as other feed ingredients such as feathermeal. In our opinion adjustments of the water treatment process (Good Manufacturing Practices), such as minimizing the time between water collection, water treatment, and handling of the sludge, and a conscious choice for the application/concentration of flocculants will be necessary in the future to yield a valuable and safe feed constituent.

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## LITERATURE CITED

Barker, H. A. Fermentation of nitrogenous compounds. In *The Bacteria, a Treatise of Structure and Function Vol. II: Metabolism*, 3rd ed.; Gunsalus, I. C., Stanier, R. Y., Eds.; Academic Press: New York, 1965; pp 151–208.

Bartholomew, B. A.; Berry, P. R.; Rodhouse, J. C.; Gilbert, R. J. Scombrotoxic fish poisoning in Britain: features of over 250 suspected incidents from 1976 to 1986. *Epidemiol. Infect.* **1987**, *99*, 775–782.

Behling, A. R.; Taylor, S. L. Bacterial histamine production as a function of temperature and time of incubation. *J. Food Sci.* **1982**, *47*, 1311–1314, 1317.

Belitz, H.-D.; Grosch, W. *Food Chemistry*; Belitz, H.-D., Grosch, W., Eds.; Springer Verlag: Berlin, 1986; pp 154, 491–492.

Beutling, D.; Janz, I.; Scheiber, G. Studies on the formation of histamine by microbes of importance to food hygiene. *Monatsh. Veterinaermed.* **1984**, *39*, 231–237.

Bjeldanes, L. F.; Schutz, D. E.; Morris, M. M. On the aetiology of scombroid poisoning: cadaverine potentiation of histamine toxicity in the guinea pig. *Food Cosmet. Toxicol.* **1978**, *167*, 157–159.

Black, M. D.; Rolow, A. M.; Nelson, C. E. Stabilization of dissolved air flotation (DAF) residue from autooxidation. In *Proceedings, XIX World's Poultry Congress*, Amsterdam, Sept 20–24, 1992; World's Poultry Science Association: Aylesbury, U.K., 1992; pp 305–307.

de Boer, E. Formation of biogenic amines in food by bacterial decarboxylases. *Ware(n)chemicus* **1988**, *18*, 67–75.

DPBF. *Diervoederwetgeving in Nederland*, deel 1; Dutch Productboard for Feedstuffs (Produktschap voor Veevoeder): The Hague, 1992; p 80; ISSN 0924-6452.

El Boushy, A. R. Y.; van der Poel, A. F. B. Protein recovery from wastewater in poultry processing plants. In *Poultry Feed from Waste—Processing and Use;* El Boushy, A. R. Y., Van der Poel, A. F. B., Eds.; Chapman and Hall: London, 1994; pp 83–98.

El Boushy, A. R. Y.; Roodbeen, A. E.; Hopman, L. C. C. A preliminary study of the suitability of dehydrated poultry slaughterhouse wastewater as a constituent of broiler feeds. *Agric. Wastes* **1984**, *10*, 313–318.

Fransen, N. G.; Van den Elzen, A. M. G.; Urlings, H. A. P.; Bijker, P. G. H.; Van Logtestijn, J. G. Pathogenic microorganisms in slaughterhouse sludge. *J. Appl. Bacteriol.* **1995**, submitted for publication.

Grant, R. A. Protein recovery from meat, poultry and fish processing plants. In *Food from Waste*; Birch, G. G., Parker, K. J., Worgan, J. T., Eds.; Applied Science Publishers: London, 1976; pp 205–220.

Heddle, J. F. Activated sludge treatment of slaughterhouse wastes with protein recovery. *Water Res.* **1979**, *13*, 581–584.

Heusinkveld, W. J. Drainage and treatment of wastewater from slaughterhouses and meat industry. *Voedingsmiddelentechnologie* **1989**, *22*, 29–31.

Ingle, D. J.; Williams-Ashman, H. G. Toxicity of ammonium acetate in rats; comparison in normal and partially hepatectomized rats. *Arch. Pathol.* **1962**, *73*, 343–351.

Kavanagh, B. V.; Moodie, S. P.; Herbert, L. S. The use of waste abattoir sludge as animal feed supplements. *Proceedings of the Sixth National Chemical Engineering Conference*, Nov 6–8 1978, Queensland, Australia; p 176.

Kerstholt, H.; Nijboer, L.; Vlam E. J. *Ware(n)Chemicus* **1983**, *13*, 66–75.

Kessels, B. G. F.; Wensing, T. Effects of environmental cadmium pollution in fattening of veal calves. *Bull. Environ. Contam. Toxicol.* **1993**, *50*, 561–564.

Lindgren, S.; Pleje, M. Silage fermentation of fish or fish waste products with lactic acid bacteria. *J. Sci. Food Agric.* **1983**, *34*, 1057–1067.

Meister, A. *Biochemistry of the Amino Acids*, 2nd ed.; Academic Press: New York, 1965.

Mulder, R. W. A. W.; Bolder, N. M.; Bisalsky, A. J. N.; Janssen, W. M. M. A. The use of flocculated sludge in poultry feeds; chemical and microbiological composition. *Tijdschr. Diergeneeskd.* **1986**, *111*, 973–978.

Norberg, A. B.; Persson, H. Accumulation of heavy-metal ions by Zoogloea ramigera. *Biotechnol. Bioeng.* **1984**, *26*, 239–246.

NRC. Nutrient requirements of poultry. *Nutrient Requirements of Domestic Animals*, 8th revised ed.; Subcommittee on Poultry Nutrition, Committee on Animal Nutrition, Board on Agriculture, National Research Council; National Academy Press: Washington, DC, 1984.

NRC. Nutrient requirements of swine. *Nutrient Requirements of Domestic Animals*, 9th revised ed.; Subcommittee on Swine Nutrition, Committee on Animal Nutrition, Board on Agriculture, National Research Council; National Academy Press: Washington, DC, 1988.

NSI. *Determination of Moisture*; NEN-ISO 1442, Netherlands Standardisation Institute: Delft, The Netherlands, 1973a; NEN-ISO 1442.

NSI. *Meat and meat products. Determination of free fat content*; Netherlands Standardisation Institute: Delft, The Netherlands, 1973b; NEN-ISO 1444.

NSI. *Meat and meat products. Determination of nitrogen content*; Netherlands Standardisation Institute: Delft, The Netherlands, 1975; NEN-ISO 937.

NSI. *Meat and meat products. Determination of ash*; Netherlands Standardisation Institute: Delft, The Netherlands, 1978; NEN-ISO 936.

NSI. *Test methods for vegetable and animal oils and fats. Determination of acidity (acid value).* Netherlands Standardisation Institute: Delft, The Netherlands, 1980a; NEN 6332 (IUPAC 2.201).

NSI. *Test methods for vegetable and animal oils and fats. Determination of peroxide value*; Netherlands Standardisation Institute: Delft, The Netherlands, 1980b; NEN 6343 (ISO 3969).

Oliver, B. G.; Cosgrove, E. G. The efficiency of heavy metal removal by a conventional activated sludge treatment plant. *Water Res.* **1974**, *8*, 869–874.

Rossin, A. C.; Sterrit, R. M.; Lester, J. N. The influence of process parameters on the removal of heavy metals in activated sludge. *Water Air Soil Pollut.* **1982**, *17*, 185–198.

Sterrit, R. M.; Lester, J. N. The influence of sludge age on heavy metal removal in the activated sludge process. *Water Res.* **1981**, *15*, 59–65.

Taylor, S. L. Histamine food poisoning: toxicology and clinical aspects. *CRC Crit. Rev. Toxicol.* **1986**, *17*, 91–128.

Urlings, H. A. P.; Fransen, N. G.; Bijker, P. G. H.; Van Logtestijn, J. G. Proteolysis and amino acid breakdown of heated and irradiated poultry byproducts and muscle tissue. *J. Anim. Sci.* **1993**, *71*, 2432–2438.

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