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FEASIBILITY OF PROTEIN RECOVERY FROM ANIMAL WASTES**

Two areas of protein recovery from large pig farm wastewaters are discussed: direct recovery of solids — screenings and indirect recovery through single cell biomass growth. The screenings should be ensilaged prior to refeeding. The SCP biomass has to be concentrated and usually dried prior to addition to fodder. From the nutrition value standpoint — yeasts and excess activated sludge are the best although yeasts have to be supplemented with carbohydrates which makes the process uneconomical. Algae have good nutritional potential, however, harvesting problems and low cell walls digestibility hinder wider application.

1. INTRODUCTION

The protein deficit in the developed countries is directly related to the deficit of fodder. In Poland close to 85% of protein is used as fodder and only 15% for direct consumption, while for 1990 the forecast is 1.1 mln tons for direct consumption and 7.7 mln tons as animal feeds. The conversion of plant protein to animal protein involves significant costs and high losses which may amount to over 80% of the applied fodder.

The increasing fodder deficit in animal husbandry can be partly alleviated in conjunction with measures leading to improvement of the wastewater treatment efficiences. Two methods of utilization of the energetic potential of animal wastes are used: 1. direct recovery and recycling of manure into fodder and 2. indirect method of conversion into microorganisms biomass — the single cell protein or SCP. The literature perusal reflects the increasing interest particularly in the second method [6, 14, 17, 19, 47].

The content of protein in various materials obtained directly or indirectly from effluents from a large industrial scale piggery, housing 10.5 thousand animals in stands without

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bedding, is presented in tab. 1. The paper will discuss the fodder values of screenings separated from piggery wastes, and of excess activated sludge, yeasts of the *Candida* type, and algae grown on treated effluents. Yields and effects will be presented and amino acids content will be defined and compared with other fodder components. Possible full scale application feasibility will be reviewed.

Table 1

Total and digestible protein content in piggery effluent waste materials
Zawartość białka ogólnego i strawialnego w odpadach z ferm trzody chlewnej

Material	Protein [mg/g DM*]		Digestibility index
	Total	Digestible	
Solids from dynamic screens	84.2	37.0	45.8
	72.2	33.7	44.2
Excess activated sludge	413.4	251.5	60.8
	376.2	237.4	63.0
Mixed algal cultures —	225.3	110.5	49.0
Chlorella sp. and *Scenedesmus*	372.1	191.4	49.9
sp. predominant	269.1	133.7	49.7
Yeasts monocultures:			
Candida robusta 1	429.2	271.0	63.1
Candida tropicalis 11	437.5	281.1	64.2
Candida utilis 3	433.0	262.7	60.5
Lemna minor from the algal ponds	274.6	216.0	78.7
*Daphnia magna*** on pig manure diluted 1 : 1	500	—	—

* DM – dry matter, ** based on data from literature [23].

The estimate of total protein (TP) content was made on the basis of total Kjeldahl nitrogen (TKN) analysis and calculation of TP = 6.25 TKN. The digestible protein (DP) content was based on the same TKN analysis made on material after 48 hours digestion with pepsin in presence of hydrochloric acid [16]. The ratio of DP/TP is equal to digestibility index (DI). The amino acids content was determined by means of amino acids analyzer AAA HD 1200E (CSRS).

2. DIRECT PROTEIN RECOVERY

Recycling of solids from piggery wastes has been practiced on experimental scale in several countries and is still not sanctioned by the Polish health authorities or the US Food and Drug Administration. Several studies have shown negative effects — notably the one by HARMON [18] who noted weight loss in swine fed anaerobically predigested solids.

In one Polish study [30] 64% and 50% of dried solids were added to the following respective fodder components in two mixes: 22% and 34% of barley, 5% and 7% of beans, 4% of corn, molasses and additional minerals. In the first case the fodder gave a yield of 865 g LW/d, while in case of the second fodder the yield was 1 019 g LW/d of cattle. The studies of meat quality revealed no significant difference between the cattle fed rations with wastes and the controls [30].

The protein and amino acids analyses of screenings have shown that they can serve as a volumetric fodder ingredient for ruminants. The screenings contain 7 to 9% total protein of 45% digestibility, large amounts of amino acids and 200 g DM (dry mass)/kg of mineral components. The low digestibility is the result of fiber content.

An interesting experiment is presently run at plant A. The screenings are placed in 20-40 cm layers on foil. The seeds of oats, rape and lupin are planted and the resulting forage, together with the base substrate, undergoes ensiling or goes to the drier. The procedure should allow for 3 to 4 crops/year provided they are covered in the winter time [54].

In summary, the difficulties in separating feces (in one case in Holland nets are used under slatted floors [37]) and low protein content of recovered screenings make direct recovery questionable economically. The competitive method for these solids woud be composting and agricultural utilization. Nevertheless, further studies should be conducted on hygienics and economics of ensiling these solid wastes, as other methods of direct recovery have proved to be inefficient.

3. CONVERSION INTO BACTERIAL SCP

Excess activated sludge contains significant quantities of protein from 380 to 530 g/kg DM [29, 40] and beneficial quantities of vitamins and minerals. Several attemps on direct refeeding of activated sludge from under–floor–oxidation–ditch back to swine were shown successful by Harmon [18].

In this country centrifuging was tried in two cases [21, 29]. In experiments in Kołbacz 22% DM was attained with 24% raw protein in the DM. The excess sludge was from the 48 h aeration tank operating at $F/M = 0.5$ kg O_2/kg MLSS d (BOD_5); the treatment system Gi-Gi is described by authors elsewhere [39]. The centrifuge cake was used for preparing silage of the composition as in tab. 2. The prepared silage had good taste and odor and was only 9% less digestible than the full silage. The first silage contained protein 36 g/kg and 440 g DM/kg while the second, respectively, 40 g/kg and 430 g DM/kg.

In the authors' experiments, excess sludge from plant A of the Vidus type (described in detail in [38]) was centrifuged in pilot scale in 5 m^3/h installation of a decanting Humboldt-Vedag centrifuge. Various coagulant aids were used to improve the dewaterability of excess activated sludge; some 13% of total solids (DM) were attained in the cake with some 39% of crude protein in the DM. The digestible protein was 237 to 251 g/kg (i.e. 61-63% DI — tab. 1). The material thus recovered was fed into an industrial dryer. Strong odours have precluded longer experiments, however, the final pelleted product had very good organoleptic properties.

Table 2

The composition of silage prepared with excess activated sludge (after [29])
Skład kiszonek przygotowanych z użyciem nadmiernego osadu czynnego (według (29])

Components	% weight	
	Silage I	Silage II
Corn grain	27	—
Barley grain	—	25
Hay cut	7	—
Straw cut	—	6
Grass silage	23	28
Activated sludge cake	43	41
Total	100	100

4. CONVERSION INTO ALGAL PROTEIN

The authors' experiments have shown that the protein content and yields (mass/area) of algae grown on piggery wastes are many times higher than any of the traditional field crops. The tests were conducted in flow-through series of four laboratory ponds 30 cm deep at 3°C-22°C temperature and with round-the clock lighting.

Both biologically treated and raw piggery wastes were used. In the raw wastes experiment the first pond was anaerobic, the second was aerated, while the remaining two have served the function of algal ponds.

Out of the various species tried only *Chlorella vulgaris* and *Scenedesmus* sp. favoured the piggery wastes environment. The biomass was harvested by sieving then was centrifuged and dried in 55°C. The average yields are depicted in tab. 3. The harvested biomass

Table 3

Algae biomass yields in experimental ponds fed with biologically treated piggery wastes
Średni przyrost biomasy w doświadczalnych stawach glonowych zasilanych gnojowicą oczyszczoną biologicznie

Retention	Biomass yield	
[days]	[g/dm^3·d]	[g/m^3·d]
9 and 12	0.04-0.05	2-2.5
15 and 18	0.17-0.27	8.5-13.5
21 and 24	0.05-0.12	3.5-6

contained 22.5 to 37.2% of protein (with DI = 49%), and 10-17% of raw cellulose, and 3-8% of fat [7].

Biomass harvesting and the problem of breaking the hardly digestible cell walls are presently the two major obstacles in fuller application of algal ponds [1, 5, 40]. Comparison of sedimentation, filtration and centrifuging has proved the latter to be the preferred method — although is still far from satisfactory.

The cellulose and hemicellulose which make up some 50% of the cell walls mass are only partly digested by ruminants. Thus the final applicability of algae as fodder can be evaluated only after a thorough disintegration of the cell walls and on samples that are free from bacteria.

The studies conducted in field scale by de PAUW have yielded similar results. De PAUW has attained [10] yields of 1-10 g DM/m^2·d at various temperatures in different seasons and has managed to grow on raw and pretreated piggery wastes *Scenedesmus* sp., *Chlorella vulgaris* and *Coelastrum probascid.* Other researchers, e.g. MÜNTZ [33] and GOLDMAN [12], have also found cell wall digestibility to be the major obstacle. The future of algae application lies in solving the two problems: separation and digestibility.

Based on present difficulties the solution is to close the algal ponds in natural ecosystems of aquaculture and using the developed biomass for fish production. Other methods to be evaluated include the use of algae as supplement to biogas generation and the development of mutants with low-cellulose-walls or larger volume to cell wall surface area ratio.

4. CONVERSION INTO YEAST PROTEIN

The production of protein by yeast is over two hundred times faster than in the conventional agriculture supplying the farms, and the studies [35, 36, 40] show that higher protein contents are attained in yeasts than in field crops.

The fermentations in authors' work were conducted in batch and semicontinuous modes on four species of the *Candida* kind: *C. tropicalis 8*, *C. tropicalis 11*, *C. robusta* and *C. utilis 3*. The species were selected in batch tests. The tests were conducted on both raw piggery wastes from a large farm of the Agrokomplex technology or on raw wastes enriched with either sucrose or beet molasses; the results are presented in tab. 4.

In the raw wastes fermentation the specific growth rate, productivity and the level of C, N, P nutrients utilization were unsatisfactory. Much higher yeast production was attained on enriched wastes, however, then the cost was comparable to yeast production from molasses alone. The yeasts contain 43-47% of crude protein of 60-64% DI. The results indicate the technological feasibility of protein recovery through yeast fermentation.

The high costs of yeasts fermentation are due to the use of an expensive and not readily available molasses and suggest the use of integrated treatment–recovery systems. The combined system of fodder yeast production, animal husbandry and a joint water–wastewater management and treatment systems will allow the decrease of the overall costs.

It should be pointed out that a lot needs to be done to increase the overall efficiency

Table 4

Technological parameters of yeast production on dilute hog farm wastes — semicontinuous tests in DO-controlled substrate addition

Wskaźniki technologiczne hodowli drożdży na podłożu gnojowicy z przemysłowych ferm trzody chlewnej w układzie z ciągłym doprowadzaniem substratu (w urządzeniu DO-STAT)

Parameter	Unit	*C. tropicalis 8*	*C. tropicalis 11*	*C. robusta*	*C. utilis 3*
		Yeasts on piggery wastes without carbon enrichment			
Maximum specific growth rate	h^{-1}	0.047	0.105	0.099	0.125
Productivity	$g\ DM/dm^3 \cdot h$	0.192	0.325	0.265	0.507
		Yeasts on wastes enriched with molasses* or sacharose**			
Maximum specific growth rate	h^{-1}	0.052	0.189	0.083	0.172
Productivity	$g\ DM/dm^3 \cdot h$	0.213	0.934	0.260	1.578

* *C. utilis*, ** *C. tropicalis 8*, *C. tropicalis 11*, and *C. robusta*.

of the process. In particular mixed yeast cultures should be considered, the mastering of the principle of continuous fermentation, and the decrease of costs of additional carbon.

Preliminary trials [40] have verified the possibility of applying the continuous process. The maintenance of the steady state for prolonged periods of time without the need for inocculum and culture replacement is of major importance to process cost decrease.

The dilute pig wastes from industrial farms (COD-non filtered equal to 6-15 000 mg O_2/dm^3) have very low easiely available carbon content. There are substitute waste materials that need to be tested, such as fatty acids, aldehydes, used oils and other agricultural wastes, as well as other yeast species. *Hansenula* kind yeast were tried and they were found to utilize the piggery substrate better due to their lower carbon requirements [40].

It seems that before the solutions to these problems are found yeast protein from piggery wastes will be economically unfeasible until the price of protein increases.

6. NUTRITIONAL VALUE OF RECOVERED PROTEIN

The amino acids content of the recovered protein is presented in tab. 5, against the reference amino acids composition of chicken egg and Food and Agricultural Organization (FAO) of UNO reference standard.

All recovered protein may be used only as fodder supplement, up to a maximum of 50% due to low digestibility. Activated sludge and yeasts have proved to be the most easiely digestible source of single cell protein (SCP), cellulolytic material being responsible for low digestibility of other SCP sources — notably algae.

The amino acids content of algae is 162-145 mg/g DM, activated sludge 265 mg/g DM and yeasts 246 to 291 mg/g DM. When calculating these amounts in terms of mass per 100 g of protein the amino acids content is 64-74 g for algae, yeast and activated sludge and some 50 g for solid screenings. The nutritional value of these proteins was determined as algae — 29%, activated sludge — 38%, yeast — 29 to 32%, and screenings only 10%.

In all cases methionine is the limiting amino acid, however, it is an inexpensive easiely available component. Although thyrosine, valine and lysine may demonstrate lower levels; the recovered SCP does contain the full spectrum of exogennic amino acids and as such is considered fit for fodder use.

The possibility of using yeasts and algae has been demonstrated experimentally [27, 40]. It seems that yeasts can be used in fodder up to 20% by fattening swine, up to 15% by small piglets and 10% by sows and boars. In feeding poultry (layers and cocks) can use up to 20% of SCP yeasts.

The experiments have documented the beneficial role of the vitamins, minerals and carbohydrates in algae, which when fed to swine (*Chlorella* sp.) up 25% in the diet resulted in better weight gains [2].

6. DISCUSSION AND CONCLUSIONS

In ideal conditions 1 000 kg LW bull produces 1 kg of protein/d, 1 000 kg soybean yield 100 kg of protein/d, 1 000 kg yeasts (DM) yield 100 000 kg of protein/d, and 1 000 kg of bacteria may yield 100 000 000 000 kg of protein/d. This short comparison [4] demonstrates the nutritional potential of SCP as the substitute of relatively inefficient other agricultural products.

The single cell protein (SCP) is synthesized in numerous countries from derived hydrocarbons. Contrary to those sources, where the danger of introducing carcinogens is quite apparent, the use of agricultural by-products for SCP synthesis has to receive a much wider attention. The energetic potential of animal wastes is 30% of the input fodder — sometimes in an unchanged form.

The work on direct recycle of piggery wastes screenings has demonstrated that both sheep and cattle can take up to 50% of dried solids in their diet. The feed containing 50% of screenings had 23% of protein, 4.23% of fat, 18.71% of fiber, and 33.37% of non-nitrogeneous substances yielded a daily gain of 1 326 g in beef cattle [11], without apparent sign of health or meat quality deterioration.

Numerous problems need to be solved before wider application of recycle and SCP conversion will be practiced. It has already been demonstrated for instance that anaerobically digested dried pig wastes sludge yields definitely negative effects when fed to swine.

Table 5

Amino acids content of the fodder protein recovered from dilute piggery wastewaters from large farms
Skład aminokwasowy białka produktów paszowych uzyskanych na bazie gnojowicy z przemysłowych ferm trzody chlewnej

Amino acids mg/g DM protein	Mixed algal cultures (*Chlorella* sp. *Scenedesmus* sp.)			Excess activated sludge		Solid screenings		Yeasts monocultures of *Candida* kind				Chicken egg	FAO
								C. robusta	*C. tropicalis 11*	*C. tropicalis 8*	*C. utilis 3*		
Lisine	3.93	3.54	4.87	4.19	4.90	3.48	3.28	4.89	5.47	5.51	5.08	6.3	4.20
Histidine	1.76	1.66	1.63	1.99	2.00	1.65	1.51	3.21	2.17	1.77	1.84	2.1	—
Arginine	3.90	4.05	3.38	3.57	3.40	3.21	3.17	2.96	3.42	2.79	2.73	6.4	—
Asparg. acid	7.46	3.18	7.17	7.06	6.57	5.23	5.27	7.09	6.79	8.07	10.81		
Treonine	4.10	3.95	3.56	3.66	4.30	2.83	2.88	3.81	4.34	3.95	3.82	5.0	2.4
Serine	3.51	3.31	3.40	3.23	3.09	2.62	2.73	3.32	3.91	3.38	3.06		
Glutamic acid	8.20	7.90	8.13	7.56	8.15	7.85	8.39	6.81	9.77	9.20	5.32		
Glycine	4.94	7.89	4.11	4.33	5.03	3.73	4.36	2.85	3.73	3.14	2.40		
Alanine	6.68	6.85	5.52	6.19	7.73	4.06	4.45	4.73	4.86	4.53	4.22		
Valine	4.89	4.79	3.89	4.64	5.62	3.73	3.82	4.05	3.88	3.83	2.31	7.14	4.20
Methionine	0.84	0.88	0.88	1.09	1.19	0.37	0.18	0.99	0.86	0.87	0.97	3.1	2.20
Isoleucine	3.83	3.63	2.96	3.37	3.88	2.71	2.62	3.89	4.03	3.49	2.90	6.8	4.20
Leucine	6.49	6.73	5.78	5.98	6.68	4.48	4.20	4.72	5.33	5.04	4.21	9.0	4.80
Tyrozine	3.13	3.01	2.39	2.51	2.86	2.13	1.84	2.57	2.50	2.64	2.14	4.4	4.80
Phenylalanine	3.94	3.86	3.72	3.77	4.03	2.99	3.01	2.92	4.19	2.99	2.41	6.0	2.80
Total	67.61	65.85	61.39	63.14	70.45	51.07	51.73	62.84	65.23	61.22	58.86		
mg/g DM	152.34	245.02	165.17	261.06	265.07	42.99	37.34	269.73	285.40	291.24	246.68		

As a rule usually higher benefits are attained when recovered protein is fed to another group of animals, as for example with DPW (dried poultry waste) fed to cattle or swine solids fed to cows and sheep.

The conclusions may be itemized as follows:

1. There are potential possibilities of direct recovery of protein in waste solids from pig wastes. Complex zoohygienic studies are, however, needed before full application, since the present experience is short-termed and narrow in scope although very encouraging. The sterilization and/or drying is not the best method of screening separation due to cost and odor problems. The desired alternative is ensiling with other fodder. The direct recycle should be in open cycles, i.e. fed to animals of other kinds.

2. Algae SCP grown on piggery wastes are an excellent food source with high protein, vitamin and minerals content. The research should in the future solve the problem of low digestibility of cell walls and the difficulties in harvesting. At present the immediate application would be in closing the cycle through aquaculture — fish cultivation.

3. Yeasts grown on piggery wastes have quality similar to the commercial product. There are no additional cost benefits when the pig wastes are used since large quantity of molasses needs to be added.

The possible more economical alternatives are integratings waste treatment and recovery systems of yeast plant and pig farm, the use of mixed yeast cultures and the use of other inexpensive waste carbohydrate sources.

4. Perhaps the most promising techniques of piggery wastes utilization involve combined waste treatment and direct as well as SCP protein recovery in industrial complexes that attempt at closing the nutrient and food cycles: nutrients–SCP–aquatic animals–fodder farm animals.

5. Excess activated sludge is found to be of excellent fodder value, however, as the SCP source it is of minor importance since the process is uneconomical for piggery wastewaters and will have to replaced in the future by low-energy-consumption treatment systems.

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MOŻLIWOŚCI ODZYSKU BIAŁKA PASZOWEGO Z GNOJOWICY Z FERM TRZODY CHLEWNEJ

Omówiono dwa zasadnicze kierunki odzysku surowca paszowego ze ścieków: bezpośrednie wykorzystanie skratek z sit i konwersja na biomasę tzw. jednokomórkowego białka (BJK). Skratki muszą być zakiszane przed skarmianiem. Masa BJK musi być zagęszczana i zazwyczaj suszona przed dodaniem do paszy. Z żywieniowego punktu widzenia drożdże i nadmierny osad czynny okazały się najlepsze. Zastosowane drożdże szczepu *Candida* wymagają jednak dodatkowego źródła węgla węglowodanowego, co czyni proces nieekonomicznym. Glony mają wysoki potencjał odżywczy, jednak problemy zbioru glonów i niska strawność ścianek komórkowych na razie utrudnia szersze zastosowanie tego procesu konwersji zanieczyszczeń rozpuszczonych w BJK.

DIE MÖGLICHKEIT DER WIEDERGEWINNUNG DES FUTTEREIWEISSES VON DER GÜLLE AUS SCHWEINEZUCHTBETRIEBEN

Besprochen werden zwei Rückgewinnungsmethoden von Futter-Rohstoffen aus dem Abwasser: eine direkte Verfütterung von Rechen- und Siebgut sowie eine Konversion zum sog. Monozell-Eiweiß (MZE). Das Siebgut muß vor dem Verfüttern eingesäuert werden. Die MZE-Futtermasse muß eingedickt und vor dem Zugeben zum Futter noch vorgetrocknet werden. Beste Nährwerte des Futters erzielt man aus Hefen und dem Überschußschlamm. Der Hefeart *Candida* muß Kohlenstoff aus Kohlenwasserstoffen zugesetzt werden was jedoch unwirtschaftlich ist. Ein hohes Nährpotential weisen Algen auf, aber es entstehen gewisse Probleme bei der Einsammlung. Schwer verdaulich ist auch die Zellwand der Algen, was einer Verbreitung dieses Konversionverfahrens der Verschmutzungen zu MZE zur Zeit noch im Wege steht.

ВОЗМОЖНОСТИ РЕГЕНЕРАЦИИ КОРМОВОГО БЕЛКА ИЗ НАВОЗНОЙ ЖИЖИ ОТ СВИНОВОДЧЕСКОЙ ФЕРМЫ

Обсуждены два основных направления регенерации кормового сырья из сточных вод: непосредственное использование осадок с сит и конверсия в биомассу так называемого одноклеточного белка (ОКБ). Осадки должны перед скармливанием заквашиваться. Масса ОКБ должна сгущаться и обычно перед добавлением в корм сушиться. С питательной точки зрения наилучшими оказались дрожжи и излишний активный осадок. Однако применение плёнчатых дрожжей требует дополнительного источника углеводного углеводорода, что делает процесс неэкономическим. Водоросли имеют высокий питательный потенциал, оданко проблемы сбора водорослей и низкая перевариваемость ячеичных стенок пока затрудняет более широкое применение этого процесса конверсии загрязнений, растворимых в ОКБ.