The Role of Trace Elements on Anaerobic Co-digestion in Biogas Production

Anthony Njuguna Matheri[1], *Member, IAENG*, Mohamed Belaid[2], *Member, IAENG*, Tumisang Seodigeng[3], Jane Catherine Ngila[4]

***Abstract*—In this study, we investigated the concentration of trace elements in the digestates in a laboratory batch anaerobic digester. Many of these trace elements are important macro and micro nutrients. The availability of these nutrients for microbes responsible for anaerobic digestion and substrate toxicity have to be controlled in biogas production. The analyzed substrates were characterized at various concentrations in the following trace elements; potassium, phosphorus, manganese, copper, calcium, molybdenum, zinc, cobalt, iron, aluminum, silver, nickel and cadmium. Trace elements like copper, silver, nickel, cadmium, zinc have been reported to be inhibitory and toxic under certain conditions in biochemical reaction depending on their concentrations. These trace elements lower biogas production above threshold concentration due to accumulation of organic acid as a result of methanogenic bacterial inhibition. There was no deficit of nutrients detected in the anaerobic digesters analysis.**

***Keywords*— Anaerobic digestion, Co-digestion, Mesophilic Temperature, Trace Elements**

Manuscript received April 13, 2016; revised April 2016. This work was supported in part by University of Johannesburg, Process Energy Environmental station (PEETS) and City of Johannesburg South Africa.

Anthony Njuguna Matheri; is with Department of Chemical Engineering; University of Johannesburg; Doornfortein, Johannesburg 2028 South Africa; Cell: +27616986335 (tonynjuguna22@gmail.com)

Mohamed Belaid; Senior lecturer and HOD at Department of Chemical Engineering; University of Johannesburg; Doornfortein, Johannesburg 2028 South Africa; Tel: +27115596402 (mbelaid@uj.ac.za)

Tumisang Seodigeng; Senior lecturer at Department of Chemical Engineering; Vaal University of Technology; Private Bag X021-Vanderbijilpark-1911, Andries Potgieter Blvd South Africa; Tel: +27169509734 (tumisangs@vut.ac.za)

Jane Catherine Ngila; Professor and HOD at Department of Analytical Chemistry; University of Johannesburg; P.O. Box 17011, Doornfortein, Johannesburg 2028 South Africa; Tel: 27115596169 (jcngila@uj.ac.za)

I. INTRODUCTION

THE economic development of developed and developing countries depend on the large extent on power generation.

With the fast depletion of the scarce non-renewable energy sources such as fossil fuel and coal which has led to human health problems, environmental degradation and major factor of the global climate change scenario, the commercial production of biogas and other alternative energy sources such as solar energy, wind energy, hydropower, geothermal will definitely give a good drive for the development of the economy. Biogas is used in the form of fuel, electricity and heat. It is desirable to create a sustainable world class energy system which has zero carbon emissions [1-3]. Biomass fermentation increases biogas demand as a renewable energy and resolves waste management problems. The most popular types of substrates are animal manure, food waste, municipal sewage sludge, vegetable, fruit waste and municipal solid waste. Municipal solid waste, food waste and vegetable/fruit waste generate more methane compared to municipal sewage sludge and animal waste thus the need for co-digestion to increase efficiency of biogas production [4]. There is high increasing energy crops use as substrate in biogas production which run into deficit trace elements with regards to manures from cattle, chicken or pig. However, variety of organic waste such as food waste, abattoir waste, kitchen waste are assumed to supply sufficient concentration and quantities of micro nutrients [5]. The co-digestion of manures and other substrates increase carbon to nitrogen (C/N) ratio and concentration of micro and macro nutrients that leads to increase in biogas production [6-8].

Biogas is a mixture of gases such as methane, carbon dioxide, hydrogen sulphide, ammonia and trace amounts of oxygen, carbon monoxide and hydrogen. It is produced by break down of organic materials using microbial under controlled conditions (parameters). The organic source is composed of biodegradables such as municipal solid waste, agricultural waste, industrial waste and animal waste [9, 10].

Anaerobic digestion of organic material takes place in series of four fundamentals steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis. Fig. 1 shows degradation steps of anaerobic digestion process. In hydrolysis step, large organic polymers such as fats, proteins and carbohydrates are broken into fatty acids, amino acids and simple sugar respectively. This step is carried out by bactericides.

Hydrolysis is followed by acidogenesis where low alcohol and organic acids are produced through fermentation process utilized by fermentative bacteria. This includes volatile fatty acids (acetic acid, butyric acid and propionic acid), gases like carbon dioxide, ammonia and hydrogen and aldehydes. In the third step (acetogenesis), the products of acidogenesis are converted to acetate, carbon dioxide and hydrogen by acetogenic bacteria. Methanogenesis is the final stage whereby methanogenes bacteria converts hydrogen, acetic acid and carbon dioxide to methane and carbon dioxide [11-13].

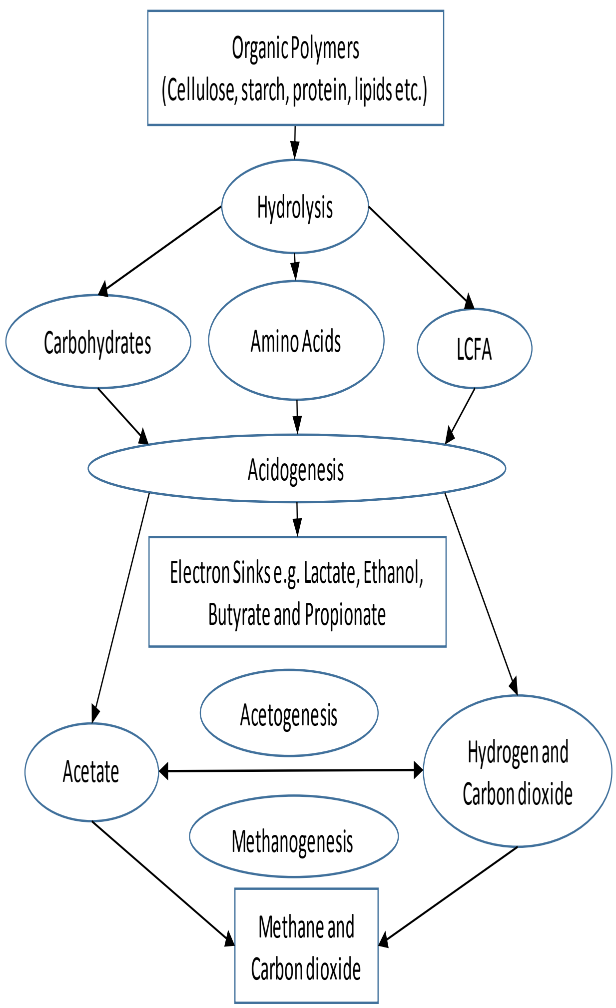

Fig. 1. Degradation steps of anaerobic digestion process.

The effective production of biogas requires optimal: temperature, partial pressure, pH, hydraulic retention time, nature of substrate, C/N ratio, stirring intensity, trace elements concentration, microbes balance and digester size [4, 14].

Methane formation by methanogens is the necessary process stage because the bacteria responsible for the process are inhibited by their own products [15]. Biogas yield is determined by the content of micro and macro elements which are responsible of methanogenesis and metabolic processes. Trace elements can be inhibitory, stimulatory or even toxic in the digestates depending on the concentration [13, 16, 17]. These co-factors in enzymes decompose larger organics to smaller molecules [4]. The most important trace elements in micro nutrients mostly involved in anaerobic digestion efficiency are cobalt, nickel, molybdenum and selenium. These elements are always supplied with feedstock and their deficiency leads to poor performance of anaerobic digestion [3, 5]. The investigated micro nutrients composed of zinc, molybdenum, manganese, copper, nickel and cobalt. The macro nutrients composed of potassium, calcium and iron were investigated. The negative influence of trace elements is determined by concentration in the substrate and pH of digestates. The fermentation under presence of trace elements are effective under high pH [4, 18, 19]. Toxic elements such as cadmium, phosphorus, chromium and lead can reduce efficiency and amount of methane formation. Toxic effect of trace metals is mostly attributed by replacing naturally occurring elements with enzyme prosthetic groups or due to disruption of enzyme function and restructure by bindings of trace metals with thiols and other groups on protein molecules [18]. Selected elements that speed up metabolism of biogas formation include potassium, calcium, magnesium, zinc, iron, zinc, cobalt and copper, but their concentration cannot exceed the threshold standards [4]. Equation 1 shows a simplified generic of anaerobic digestion process [20].

$$C_6H_{12}O_6 \rightarrow 3CO_2 + 3CH_4 \quad (1)$$

Previous work done on the trace metals concentration has been reported by [5, 21, 22] as shown in Table I.

Justification by knowledge of importance and general functions of selected trace elements in various general metabolism processes in methanogenesis are shown in Table II. These elements are involved in decomposition of numerous enzymes compounds to smaller molecules [5].

The main objective of this study was to investigate the concentrations of trace metals in the digestates and their influence in biogas production. It is important to determine appropriate concentration of trace metals required as this determines supplement required in the substrates and in management of liquid and solids residual utilization and disposal [23].

TABLE I
RECOMMENDED CONCENTRATION OF TRACE ELEMENTS IN ANAEROBIC DIGESTION [5].

		Weiland (2006)	Bischofsberger (2005)	Mudrack and kunst (2003)	Seyfried at al. (1990)	Takashima Speece (1990)
Element	Symbol	Concentration (mg/l)				
Boron	B	-	-	-	-	-
Calcium	Ca	-	-	-	-	>0.54-40
Cobalt	Co	0.003-0.06	0.06	0.003-0.06	0.003-0.06	>0.00059-0.12
Chromium	Cr	-	0.005-50	-	-	-
Copper	Cu	-	-	-	-	-
Iron	Fe	1-10	-	1-10	1-10	>0.28-50.4
Magnesium	Mg	-	0.005-50	-	-	360-4800
Manganese	Mn	-	0.005-50	-	-	-
Molybdenum	Mo	0.005-0.05	0.05	0.005-0.05	0.005-0.05	>0.00096-0.048
Nickel	Ni	0.005-0.5	0.006	0.005-0.5	0.005-0.5	0.0059-5
Lead	Pb	-	0.02-200	-	-	-
Sulphur	S	-	-	-	-	0.32-13000
Selenium	Se	-	0.008	-	0.008	0.079-0.79
Tungsten	W	-	-	-	0.1-0.4	0.018-18.3
Zinc	Zn	-	-	-	-	-

TABLE II
THE IMPORTANCE AND FUNCTIONS OF TRACE ELEMENTS [5].

Element	General function (microorganisms)	Element	General function (microorganisms)
Boron	➢ cofactor of enzymes	Manganese	➢ activate enzymes of bacteria
Calcium	➢ membrane permeability ➢ influence toxic effects of other metals ➢ enhance toxic effects of other metals		➢ stabilizes methane producing bacteria ➢ redox reaction ➢ cofactor of various enzymes
Chromium	➢ glucose metabolism	Molybdenum	➢ inhibitor of sulphate reducing bacteria
Cobalt	➢ metallic enzyme activator		➢ cofactor of various enzymes
	➢ can inhibit metabolism	Nickel	➢ synthesis of coenzymes
Copper	➢ metallic enzyme activator		➢ cofactor of urease
	➢ can inhibit metabolism	Selenium	➢ hydrogenase in methane producing bacteria
	➢ reduce other metals toxicity	Sulphur	➢ cofactor and components of many proteins and metabolic compounds
	➢ pigment	Zinc	➢ metallic enzyme activator
Iron	➢ redox property		➢ stimulates cell growth
	➢ electron acceptor		➢ can exacerbate toxic effects of other metals and inhibit metabolism
Magnesium	➢ enzyme activator		➢ hydrogenase in methane producing bacteria

ISBN: 978-988-14048-0-0
ISSN: 2078-0958 (Print); ISSN: 2078-0966 (Online)

II. MATERIAL AND METHODS

Cow dung, pig waste, chicken droppings and grass clippings were collected at the farm in Gauteng Province. Waste characterization was done to ascertain the composition. This included physical and chemical composition with regards to C/N ratio, volatile solids, total solids and elemental analysis for carbon, nitrogen, sulphur, hydrogen and trace elements in accordance with the standard methods (APHA 2005) [24].

To determine biogas production rate, a batch digester was fed with the co-digested substrates and inoculum under pre-set conditions of 37 ^{0}C and pH of 7 as shown in Fig. 2. The digesters were flushed with nitrogen to expel the oxygen and create an anaerobic condition. They were then immersed in the water bath and kept under the set conditions. The gas produced was measured using downwards displacement method whereas liquid samples were analyzed on a daily basis until the end of retention time in accordance with the standard methods [25]. Fig. 2 shows the biogas digestion set up.

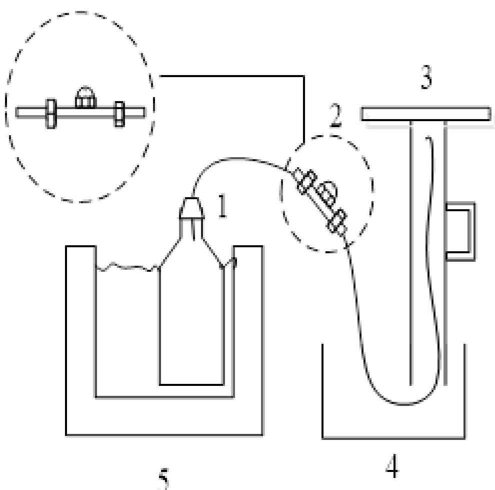

Fig. 2. Biogas Production Set-up (1 Digester, 2 T-union, 3 Measuring cylinder, 4 Water bucket, 5 Thermostatic water bath)

III. RESULTS AND DISCUSSION

In this study, cow dung, pig waste and chicken droppings all co-digested with grass clippings were evaluated for the purpose of getting the bio-methane potentials and bio-chemical kinetics at optimum temperature (37 ^{0}C) and initial pH of 7. Table III shows the substrate characterization of the substrates.

TABLE III
SUBSTRATE CHARACTERIZATION

Substrate	C	H	N	S	VS (%)	TS (%)	C/N ratio
Grass clippings	19.10	1.04	0.93	0.00	64.08	87.88	20.54
Chicken manure	63.67	0.85	3.11	2.25	11.75	18.74	20.47
Cow dung	14.87	1.65	0.84	3.66	78.72	91.55	17.70
Pig dropping	42.26	0.70	2.62	0.00	55.70	76.80	16.16

The composition of trace elements in substrates were to be determined before setting up the biogas digestion process and during digestion. This was done on substrate characterization. Maximum and minimum concentration varied with various substrates. Trace elements concentration evaluated in the digestates samples are presented in Table IV.

Trace elements such as zinc and copper, magnesium, calcium and potassium are essential for microbial growth [4]. Microorganisms are able to accumulate trace elements in them thus reducing their concentration in the digestates [26]. High level of trace elements can reduce the efficiency of biogas production. copper, cadmium, lead, nickel and zinc exert toxic effect and inhibiting anaerobic digestion [4].

TABLE IV
TRACE ELEMENTS AVERAGE ABUNDANCE

Element	Symbol	Units	Average trace metals concentration		
			Chicken dropping	Cow dung	Pig waste
Aluminium	Al	mg/l	0.13	0.04	1.01
Cadmium	Cd	mg/l	0.03	0.07	0.07
Cobalt	Co	mg/l	2.14	1.28	3.02
Copper	Cu	mg/l	4.81	2.81	7.06
Calcium	Ca	mg/l	0.79	3.04	6.86
Iron	Fe	mg/l	1.13	2.67	1.09
Manganese	Mn	mg/l	4.22	4.19	18.40
Molybdenum	Mo	mg/l	3.28	2.64	5.03
Nickel	Ni	mg/l	0.18	0.17	0.25
Phosphorus	P	mg/l	7.79	1.40	9.66
Potassium	K	mg/l	6.26	4.95	17.63
Silver	Ag	mg/l	0.60	0.37	1.16
Zinc	Zn	mg/l	1.78	1.46	3.01

Fig. 3 shows the trace elements concentration in cow dung, pig waste and chicken droppings all co-digested with grass clippings. The inference of the trace elements with regards to inhibitory, stimulatory and toxicity to anaerobic digestion included:

ISBN: 978-988-14048-0-0
ISSN: 2078-0958 (Print); ISSN: 2078-0966 (Online)

Potassium—The potassium content evaluated ranged from 6-18 mg/l. The highest potassium noted was 17.63 mg/l in the pig waste and low on the cow dung. The toxic threshold for potassium has been estimated to be 3000 mg/l [4].

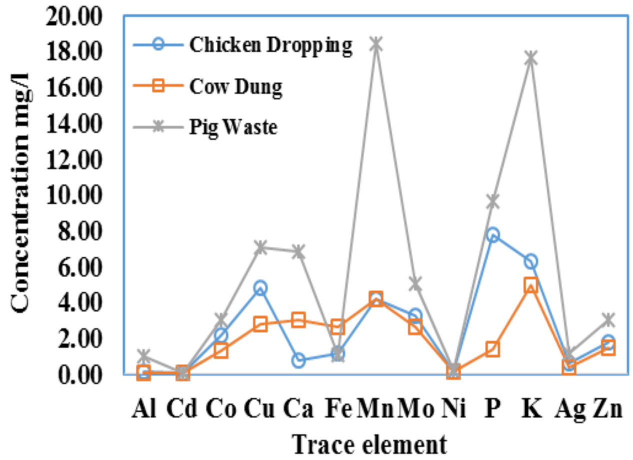

Fig. 3. Trace elements concentration in substrates

Calcium—Appropriate pH in substrate is determined by optimum content of alkaline-forming minerals as well as neutralization anions. Calcium is a pH adjusting mineral. In this study, the calcium toxic concentration was below the threshold of 2800 mg/l as specified by [4, 21].

Manganese—The concentration ranged from 4-19 mg/l. The manganese in the digestates was below recommended threshold concentration of 50 mg/l in accordance to [4]. It was the most abundant trace element in the digesters.

Phosphorus—The phosphorus concentration ranged from 1-10 mg/l. It was reported high in pig waste and low in cow dung.

Cobalt—The cobalt concentration ranged from 1-4 mg/l. It acts as metallic enzyme activator and could inhibit metabolism above threshold concentration.

Copper—The threshold for copper concentration is 400 mg/l. As seen in this study, it ranged from 2-8 mg/l. Above the threshold concentration, copper will inhibit the biogas production [4].

Molybdenum—It was found to range from 5-6 mg/l in concentration. The value was higher than the one recommended by [4].

Zinc—The zinc concentration ranged from 1-3 mg/l. It was above the recommended threshold of 1 mg/l recommended by [4]. Above threshold, zinc inhibits methane formation and enhance methane formation below threshold. Good co-digestion enhance its suitability.

Iron—The iron concentration in the digester ranged from 1-3 mg/l. The threshold of iron concentration as specified by [21] is 10 mg/l.

Nickel—The nickel concentration in the digester ranged from 0.10-0.25 mg/l. It improves methane yield and maintains process stability. The toxic threshold of nickel is reported to be 10 mg/l [4]. In our findings, the nickel was below the threshold to stimulate biogas production.

Aluminium—The aluminium concentration in the digester ranged from 0.01-1.01 mg/l. There was no recommendation found for aluminium concentration in the literature.

Silver—The silver concentration in the digester ranges from 0.06-1.16 mg/l. In the literature there was no recommendation found for silver concentration.

Cadmium—The cadmium level ranged from 0.03-0.07 mg/l. The threshold below 0.18 mg/l pose no threat to anaerobic digestion [4]. Co-digestion of the substrate enhance reduction of cadmium concentration.

According to the observation, pig waste had higher level of trace elements followed by chicken droppings and lastly cow manure on average abundance concentration. This was attributed by their feeding habit in the respective animals and their stomach compartment. Cows are ruminant's animals with four compartment stomach. Fermentation starts in their stomach and there is tendency of microbial activity leading to reduction of trace elements. Chicken and pigs have one stomach compartment. There is little fermentation process taking place in their stomach and this leads to presence of high concentration of trace elements. The abundance of trace metals did not show deficiency compared to the recommendation in the biogas production literature. Well monitored trace elements and co-digestion of the substrates enhanced process stability and improved biogas production rate.

IV. Conclusion

The concentration of trace elements investigated were within permissible limits suitable for biogas production. In general, co-digestion of organic substrate improves the abundance of trace elements thus increasing the concentration of micro and macro nutrients. The main inference from the above discussion is that trace elements maybe inhibitory, stimulatory and even toxic to anaerobic digestion, where the extent depends on elements concentration. The concentration of trace metals in the digester should be monitored under retention time to guarantee the efficiency of methane formation. Further comprehensive research to determine the optimum concentration for micro and macro nutrients and their interaction with microbial activity in anaerobic digestion is required.

Acknowledgements

The author wish to express their appreciation to Process Energy Environmental and Technology Station (PEETS) funded by South Africa National Energy Development Institute (SANEDI), City of Johannesburg (COJ) and Technology Innovation Agency (TIA) through Prof Charles Mbohwa, Chemical Engineering and Applied Chemistry Departments at the University of Johannesburg for allowing us to work in their laboratories. Dr. Robert Huberts for assisting with some of the equipment for analysis.

REFERENCES

[1] H. Allan *"Grass productivity".* Island Press Conservation Classics Series, Washington DC, 1998: p. 56-89.

[2] G.K. Latinwo and S.E. Agarry, *Modelling the Kinetics of Biogas Production from Mesophilic Anaerobic Co-Digestion of Cow Dung with Plantain Peels.* International Journal of Renewable Energy Development (IJRED), 2015. **4**(1): p. 55-63.

[3] M. Lebuhn, F. Liu, H. Heuwinkel, A.Gronauer, *Biogas production from mono-digestion of maize silage-long-term process stability and requirements.* Water science and technology, 2008. **58**(8): p. 1645.

[4] M. Bożym, I. Floozak, P. Zalanowska, J. Wojdalski, M. Klimkiewicz., et al.,*An analysis of metal concentrations in food wastes for biogas production.* Renewable Energy, 2015. **77**: p. 467-472.

[5] A. Schattauer, E. Abdoun, P. Weiland, M. Plochl, M. Heiermann.,, *Abundance of trace elements in demonstration biogas plants.* Biosystems engineering, 2011. **108**(1): p. 57-65.

[6] R. Braun, E. Brachtl, and M. Grasmug, *Codigestion of proteinaceous industrial waste.* Applied biochemistry and biotechnology, 2003. **109**(1-3): p. 139-153.

[7] P. Weiland, *Anaerobic waste digestion in Germany–Status and recent developments.* Biodegradation, 2000. **11**(6): p. 415-421.

[8] L. Zhang, Y.W. Lee, and D. Jahng, *Anaerobic co-digestion of food waste and piggery wastewater: focusing on the role of trace elements.* Bioresource technology, 2011. **102**(8): p. 5048-5059.

[9] A.S. Sambo, B. Garba, and B.G. Danshehu, *Effect of some operating parameters on biogas production rate.* Renewable Energy, 1995. **6**(3): p. 343-344.

[10] S. Dangoggo, M. Aliyu, and A. Atiku, *The effect of seeding with bacteria on biogas production rate.* Renewable energy, 1996. **9**(1): p. 1045-1048.

[11] R. Sundararajan, S. Jayanthi, and R. Elango, *Anaerobic digestion of organic fractions of municipal solid waste and domestic sewage of Coimbatore.* Indian Journal of Environmental Health, 1997. **39**(3): p. 193-196.

[12] I. Angelidaki, H.V. Hendriksen, I.M. Mathrani, J.E. Schmidt, A.H. Sorensen, B.K. Ahring., *The biogas process. Lecture notes for: Energy from biomass (6362)*. 1996.

[13] A. Mudhoo. and S. Kumar, *Effects of heavy metals as stress factors on anaerobic digestion processes and biogas production from biomass.* International Journal of Environmental Science and Technology, 2013. **10**(6): p. 1383-1398.

[14] A.N. Matheri, M. Belaid, S. Tumisang, C.J. Ngila., *The Kinetic of Biogas Rate from Cow Dung and Grass Clippings.* 7th IIENG International Conference of latest trends in Engineering and Technology (ICLTET'2015) Pretoria, South Africa, November 2015. , 2015.

[15] E. Nettmann, et al., *Polyphasic analyses of methanogenic archaeal communities in agricultural biogas plants.* Applied and environmental microbiology, 2010. **76**(8): p. 2540-2548.

[16] J.K. Oleszkiewicz and V.K. Sharma, *Stimulation and inhibition of anaerobic processes by heavy metals—a review.* Biological Wastes, 1990. **31**(1): p. 45-67.

[17] S.S Şengör., et al., *Influence of heavy metals on microbial growth kinetics including lag time: mathematical modeling and experimental verification.* Environmental Toxicology and Chemistry, 2009. **28**(10): p. 2020-2029.

[18] Y.R. Chen, J.J. Cheng, and K.S. Creamer, *Inhibition of anaerobic digestion process: a review.* Bioresource technology, 2008. **99**(10): p. 4044-4064.

[19] I.J. Kugelman and P.L. McCarty, *Cation toxicity and stimulation in anaerobic waste treatment.* Journal (Water Pollution Control Federation), 1965: p. 97-116.

[20] T.R. Sreekrishnan, S. Kohli, and V. Rana, *Enhancement of biogas production from solid substrates using different techniques—a review.* Bioresource technology, 2004. **95**(1): p. 1-10.

[21] M. Takashima, R.E. Speece, and G.F. Parkin, *Mineral requirements for methane fermentation.* Critical Reviews in Environmental Science and Technology, 1990. **19**(5): p. 465-479.

[22] P. Weiland, *Anforderungen an Pflanzen seitens des Biogasanlagenbetreibers, TLL-Jena, Eigenverlag, 12.* Thüringer Bioenergietag, 2006: p. 26-32.

[23] B. Demirel and P. Scherer, *Trace element requirements of agricultural biogas digesters during biological conversion of renewable biomass to methane.* Biomass and Bioenergy, 2011. **35**(3): p. 992-998.

[24] A.P.H.A and F.W. Environmental., *Standard methods for the examination of water and wastewater.* American Public Health Association (APHA): Washington, DC, USA, 2005.

[25] Copyright., H., *Hach standards http://www.hach.com/quick.search-download.search.jsa?keywords=METHOD%20OF%20ANALYSIS%20FOR%20METALS* Accessed 2015.

[26] L.E. De-Bashan, P. Magallon, H. Antoun, Y. Bashan., *Role of glutamate dehydrogenase and glutamine synthetase in chlorella vulgaris during assimilation of ammonium when jointly immobilized with the microalgae-growth-promoting bacterium azospirillum brasilense1.* Journal of Phycology, 2008. **44**(5): p. 1188-1196.