Physic-chemical composition of concentrated vinasse for their assessment in animal diets

Idania Scull[1], Lourdes Savón[1], Odilia Gutiérrez[1], Elaine Valiño[1], Ibett Orta[1], P.O. Mora[1], Himilce Orta[1], Yusmely Ramos[1], A. Molineda[2], G. Coto[3] and Aida Noda[1]

[1]*Instituto de Ciencia Animal, Apartado Postal 24, San José de las Lajas, Mayabeque, Cuba*
[2]*Universidad Central de las Villas "Martha Abreu", Carretera de Camajuaní km. 5,5, Santa Clara, Villa Clara, C.P. 54830*
[3]*Centro de Histoterapia Placentaria, Calle 18 No. 4302 e/ 43 y 47, Miramar, Playa, La Habana, Cuba*
Email: idascull@ica.co.cu

The physic-chemical characterization of concentrated vinasse from the Havana Club rum factory of San José de las Lajas municipality, Mayabeque province, was conducted to assess its potentialities as animal feeding and contribute, at the same time, with the environment. The vinasse samplings were conducted in the storing tanks of the Swine Unit and the Ruminants Production Center "Atrevido" from the Institute of Animal Science. The proximal analysis, pH, specific weight, reducing sugars and amino acids composition were considered for the physic-chemical composition. A descriptive analysis with five repetitions was applied for processing the data. The microbiological analysis was also carried out to know the presence of polluting microorganisms. The values of pH and specific weight of the product ranged between 3.92-4.20 and 1.08-1.13 gL^{-1}, respectively. The DM content varied between 21-29 %, while that of ash remained between 23-25 % DM and the reducing sugars fluctuated from 3.69 to 4.95 %. The mean mineral composition concentration of most elements (%) calcium, 3.54; magnesium, 1.20; phosphorous, 0.26; potassium, 6.78 and sodium, 1.71 was within the required ranges for different animal species. The CP levels oscillated between 11-14 % of DM, there was high proportion of glutamic acid, lysine and aspartic acid in the amino acidic profile. No polluting microorganisms were found in the product. The results of the physic-chemical characterization of vinasse showed the potentialities of this by-product as animal feeding. Further physiological and of productive performance studies are recommended to clear up the effects of this product on the different species and animal categories.

Key words: *vinasse, chemical composition, animal feeding, environment.*

Searching for alternative feedings for animal production, mainly with the use of available resources, is still an unsolved problem. One of the main challenges is to find nutrient sources of easy acquisition. Products like vinasse, considered as non-conventional feedings, are not universally used in animal feeding, but if they are properly used they could be an important element in the sustainable production systems (Bermúdez 1994 and Martínez *et al* 2010), in respect to their low cost and the presence of organic compounds (acids, alcohols, sugars), minerals and nitrogen compounds (amino acids, peptides). Due to these characteristics, vinasse is considered a very aggressive by-product that provokes serious environmental problems in the hydric resources of its deposition (Gálvez 2005).

Vinasse is obtained from the fermentation process of alcohol distilling, from raw materials such as molasses or juices of sugarcane, maize, beet, among others, and from the use of yeasts (Robertiello 1981 and Hernández *et al* 2008). This process occurs in a distillation column, from which a liquid without ethanol, called must or vinasse is obtained. The composition of this by-product varies according to the conditions of the fermentation and distillation process and the raw materials used. That is why its chemical assessment is so important if it is intended to be used directly in animal feeding or indirectly in the obtainment of other feeds. The technology used implies a concentration system between 32-35 °Brix and has, as special characteristic of the process, Saccharomyces, increasing the protein level.

The objective of this study was the physic-chemical characterization of the vinasse obtained from the elaboration process of Havana Club rum, in San José de las Lajas municipality, Mayabeque province, to assess its potentialities for animal feeding and eliminate the environmental pollution originated by this product.

Materials and Methods

Concentrated vinasse was used for this study, obtained from the Havana Club rum factory of the Mixed Enterprise Havana Club, located in San José de las Lajas municipality, Mayabeque province. The samples were taken from five production lots in the second semester of 2008. They were stored in tanks placed in Swine Unit (sample 1) and in "Atrevido" Unit (sample 2) of the Institute of Animal Science.

Samples taking. The sampling was randomly conducted every 10 min. The outlet faucets of the inferior part of the storing tanks were used. They were taken by quintuple and stored in crystal flasks previously sterilized. They were kept at room temperature until the analyses. The samples were conducted once every month for six months.

Chemical analysis. The bromatological analysis was conducted according to AOAC (1995). The DM was determined through the drying out method proposed in the Manual Producción Azucarera (1990). The CP was determined by the method of Bernstein, cited by

Meir (1986). The pH determinations were conducted potentiometrically. The minerals concentration was determined through atomic absorption, with a Pye Unicam SP-9 equipment.

Determination of amino acids. A part of the sample was taken for determining the amino acids and it was hydrolyzed with HCl 6 N under reflowing conditions, at 110 °C, to divide the peptidic chain and guarantee the presence of amino acids as free molecules in the solution. The analyses were conducted by ionic interchange chromatography with an auto-analyzer Alpha Plus, according to the methodology of Melgarez (1985).

Microbiological analysis. The counting of polluting microorganisms was carried out following the norms and procedures of Feeding Microbiology for human and animal consumption and is shown in table 1.

Table 1.Norms for determining polluting microorganisms

Indicators	Reference
Counting of microorganisms mL^{-1}	NC/ISO 4833-2002
Counting of total coliforms mL^{-1}	NC/ISO 4832-2002
Counting of molds and yeasts mL^{-1}	NC/ISO 7954-2002
Determination of salmonella $25mL^{-1}$	NC 605:2008

Statistical analysis. The descriptive analysis with five repetitions was used for analyzing the results through the software Infostat, version 1.0, of Balzarini *et al*. (2001).

Results and Discussion

In the samples assessed, the liquid product had particles in suspension of brown color, similar smell to that of the final molasses and malt taste.

The physic-chemical characteristics of concentrated vinasse are shown in table 2. They all had acid pH. These values are proper of vinasses obtained from sugarcane (Gómez 1996) and favor the preservation of the product, as most of the microorganisms do not grow under these pH values. This indicator is a detriment to the product reception, as the store deposits may have a rapid corrosion of the walls and of the inner structure, resulting in the crack and drainage throughout the walls and its canalization to the soil.

Water is an important component in most of the feeding products. In the case of vinasse, it varies considerable, representing between 68-80 %. The DM determination was conducted through the dissection method using a dispersing, inert and dry culture, mixed with the sample to facilitate the water evaporation. This method avoids the losses of volatile compounds, when using the temperature for drying (60ºC). The DM values found are superior to those reported for other vinasses obtained in this same province (ICIDCA 2000), due to the technology for obtaining the product that has a water extraction process that concentrates the nutrients.

The indicators of CP and TP had higher results dispersion than the rest of the indicators assessed, with superior variation coefficient (VC): sample 1(5.13, 1.18 %), sample 2 (3.05, 8.46 %). This variability could be due to the lack of samples homogenization as agitating the product for conducted the sampling is not possible. This factor should be considered in the future. The CP oscillated in a range of 11.5-14 %, while the TP represented between 72-70 % of the CP. Hernández *et al*. (2008) found inferior values of CP in the vinasse coming from cachaza (3.75 %). However, FEDNA (2003) reported superior levels in the vinasse of beet molasses (17.9 %) in respect to those found in this study. These results are relevant and confirm that previously explained about the chemical composition variability.

The determination of the protein fraction is one of the most important indicators for using vinasse as animal feeding and as raw material for elaborating new feeds (Mora *et al*. 2010). This aspect should be further studied due to the benefits of its functional diversity for the different animal species (enzymatic, of transportation and storing, mechanical motility, protection, regulator, among others).

The protein quality is an important concept in protein nutrition, mainly determined by the profile and proportion of amino acids, although other structural characteristics and solubility may affect its digestibility and, thus, its nutritional value (Millward 2004).

Table 2. Physic-chemical characterization of concentrated vinasse

Indicators	Sample 1 (n=5)	Statistics		Sample 2 (n=5)	Statistics	
		SD	VC (%)		SD	VC (%)
pH	4.08	0.07	1.92	3.92	0.008	0.21
Specific weight (%)	1.12	0.004	0.37	1.085	0.008	0.07
Dry matter (%)	29.31	0.02	0.07	21.33	0.20	0.94
OM (%)	77.01	2.71	1.21	76.37	0.19	0.26
Ash (%)	25.00	0.04	0.18	23.63	0.20	0.83
CP (%)	12.39	0.63	5.13	13.26	0.40	3.05
TP (%)	8.89	0.10	1.18	9.29	0.79	8.46
Reducing sugars (%)	4.43	0.9	1.02	4.01	1.10	0.96

Vinasse showed an amino acidic profile (table 3) coinciding with the percent value of TP. It is important to emphasize that both samples had high values of essential amino acids (leucine, isoleucine, valine, tyrosine, histidine, phenylalanine, lysine, threonine) and were comparable or superior to those of the FAO's pattern for monogastric species (D' Mello 1995). An aspect to highlight, and of great importance for monogastric species, is the presence of methionine, as the non-conventional feedings do not have it or have small amounts, although sometimes the analysis is not conducted. The results allow classifying this residue as of good biological value, so it can be used in animal feeding (Barros 2010).

The concentration of macro and micro nutrients are shown in table 4. In general, when determining the minerals, there was higher variability and less precision than in the rest of the indicators of the chemical composition, evidenced by the high VC. The effect of the sampling method on the mineral composition of vinasse should be further studied to prove if these results are due to the lack of sample homogenization.

Calcium has an important function regulating the permeability of the cell membranes, coagulating blood and activating key enzymes in the animal metabolism. For all these reasons, its counting in the feedings is essential. The literature data, in respect to this type of residue, are contradictory. Sarria and Serrano (2008) reported Ca (0.02-0.03 %) and Mg (0.01-0.02 %) concentrations in sugarcane concentrated vinasse. These values are very inferior to those found in this study. However, the data informed by Leal *et al*. (2006)

Table 3. Amino acidic composition of vinasse

Aminoacids	$g100g^{-1}$ Sample1	$g100g^{-1}$ Sample 2	$g100g^{-1}$ FAO pattern[1]
Aspartic acid	7.69	8.85	-
Threonine	5.66	7.31	2.9
Serine	4.64	4.81	-
Glutamic acid	12.11	13.23	-
Prolina	4.98	6.02	-
Glycine	4.08	3.61	-
Alanine	3.74	2.50	-
Cysteine	1.81	1.81	2.0
Valine	5.66	7.13	1.3
Methionine	4.53	nd	2.3
Isoleucine	6.79	5.76	4.3
Leucine	4.53	3.10	4.9
Tyrosine	6.23	4.99	2.9
Phenylalanine	5.09	4.73	2.9
Histidine	5.09	4.72	-
Lysine	9.96	9.28	4.9
Arginine	7.47	10.57	-

nd: not determined, 1 D´ Mello (1995)

Table 4. Mineral composition of vinasse

Minerals	Sample 1	Statistics		Sample 2	Statistics	
		SD	VC (%)		SD	VC (%)
Calcium (%)	4.06	0.14	5.32	3.02	0.16	3.57
Magnesium	1.14	0.04	3.12	1.27	0.03	3.57
Phosphorous (%)	0.24	0.01	1.03	0.28	0.04	0.98
Potassium (%)	6.36	0.06	3.83	7.20	0.28	1.08
Sodium (%)	1.58	0.14	3.01	1.84	0.05	8.72
Fe (%)	0.07	0.003	1.37	0.08	0.001	4.56
Zn(%)	0.04	0.004	32.50	0.08	0.002	11.32
Cu(ppm)	6.60	0.004	1.93	5.00	0.0009	5.39
Pb (pmm)	12.90	0.69	11.18	19.34	2.16	5.34
Co (pmm)	6.49	0.46	1.77	8.67	0.15	7.04

Table 5. Results of the microbiological analysis of vinasse

Indicators	Sample 1	Sample 2
Counting of microorganisms mL^{-1}	$< 1x10^4$	$< 1x10^4$
Counting of total coliforms mL^{-1}	$< 1x10^2$	$< 1x10^2$
Counting of molds and yeasts mL^{-1}	$< 5.7x10^4$	$< 5.1x10^4$
Determination of salmonella $25ml^{-1}$	Ausente	Ausente

in *Agave cocui* vinasse for Ca 3.1-3.9 % are similar to those of this study, while the Mg content (0.52-0.82 %) differed. Although defficient in phophorous, vinasse was rich in other mineral elements. Other studies are needed to determine and clear up the performance of heavy metals in this type of product.

The microbiological analysis (table 5) was carried out to now the presence of polluting microorganisms. Considerable amounts of yeasts were observed, in correspondence with the expectations according to the obtainment process. No polluting elements were found, maybe related with proper acidity of the residue, and did not favor the prolificacy of odd microbial agents in the product. This residue may be used for animal feeding from the healthy point of view.

The results showed the possibility of using this residue as animal feeding due to the content of essential nutrients for the metabolism. Including vinasse as a source of animal feeding is an option for sustainable development. Further studies are needed in respect to the effect of this residue on the different animal species. Deepening in the chemical composition of the product is also recommended, mainly on the organic compounds and the effects of this residue when included in the animals' diet.

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