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Review
GLYCERIN AS BYPRODUCT OF BIODIESEL PRODUCTION, ITS CHARACTERISTICS, APPLICATIONS AND SOIL USE

Revisión bibliográfica
Características de la glicerina generada en la producción de biodiesel, aplicaciones generales y su uso en el suelo

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ABSTRACT. Biofuels, such as bioethanol and biodiesel, represent attractive energy sources because they are made from renewable materials. Crude Glycerol is the major byproduct of the biodiesel industry.; about 2 million tons of glycerol consistently reaches the market every year, consequently several research efforts being made in order to find ways for utilizing glycerol. The aim of this work was to carry out an up-to-date revision of the information related to the characteristics of this glycerol and its general applications with emphasis on its use in the soil. The characterization of crude glycerol from biodiesel production, for different research worker, revealed heterogeneous results on the composed of each variable and its intervals. A total application of this glycerol that have already studied, demand a deep consideration in each characterization. Aspects like row materials employed in the biodiesel process production and methods of purification used by producers should pay particular attention. The use of glycerol seems to be a convenient way for increasing soil fertility, by increasing the amount of microorganisms; main fixed nitrogen microorganism, however, issue like the glycerol characteristic (saline contents, pH, methanol contents), and the applications frequency most be taking in to account.

Key words: organic matter, microorganisms, nitrogen fixation

RESUMEN. Los biocombustibles como el bioetanol y el biodiesel constituyen una fuente atractiva de energía porque se obtienen de recursos renovables. El principal subproducto del biodiesel es la glicerina cruda; se estima que más de dos millones de toneladas llegan al mercado cada año, por lo que la búsqueda de aplicaciones ha atrapado el interés de la comunidad científica. El objetivo del presente trabajo fue realizar una revisión actualizada de la información relacionada con las características de esta glicerina y sus aplicaciones generales con énfasis sobre su uso en el suelo, así como aportar algunos criterios y puntos de vista de los autores. La caracterización realizada por los investigadores a este subproducto, reveló resultados heterogéneos en las variables medidas y en los intervalos reportados para cada variable. Las múltiples aplicaciones de la glicerina que ya han sido estudiadas, demandan una consideración más profunda en cada caracterización. Aspectos como la materia prima empleada y proceso de producción y purificación aplicados en la obtención del biodiesel, merecen particular atención. El uso de la glicerina cruda se revela como una contribución a la fertilidad del suelo por el impacto positivo sobre el desarrollo de los microrganismos, principalmente los fijadores de nitrógeno; sin embargo, aspectos como las características de este subproducto (contenido salino, pH, contenido de metanol) y la frecuencia de aplicación, deben ser considerados.

Palabras clave: materia orgánica, microorganismos, fijación del nitrógeno

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INTRODUCTION

Biofuels are an attractive source of energy; they come from a renewable raw material and are also biodegradable, non-toxic, with fewer undesirable emissions (CO, aromatic hydrocarbons, soot particles, sulfur and nitrogen oxides and metals) (1). Its estimated world production is about 750 000 t per year and this industry continues growing spectacularly (2).

Biodiesel (biofuel) is a fatty acid alkyl-ester obtained from the esterification of animal fat or vegetable oil (triglycerides) with a short-chain alcohol (usually methanol or ethanol). The main oleaginous materials used come from the palm, rapeseed, soybean, sunflower, peanut, olive, and mustard, among others (3). To mitigate environmental effects, the use of marginal crops, such as *Jatropha curcas* (pine nut) and others has been pointed out, which do not require fertile lands, because they proliferate in arid soils with poor nutrients, high levels of radiation and low rainfall (4).

Glycerin is the main biodiesel by-product; more than two million tons are estimated to reach the market every year (5). One kg glycerin is generated per 10 kg biodiesel (6). It is considered that only a nonsignificant fraction (less than 5 000 ton) is synthetically obtained (5). Both its purification and the search for crude glycerin applications have caught the scientific community interest, due to the rapid growth of biodiesel production and its marked tendency to increase.

Those problems related to the high generation of crude glycerin can be mitigated if the results of its multiple current uses are investigated and implemented in different economy sectors. Particularly, the use of raw glycerin as organic matter added to the soil is gaining daily interest. It has been found that when crude glycerin is applied to the soil, plant growth increases and the added nitrogen is fixed as ammonium nitrate (7), so reducing the use of fertilizers and mitigating environmental pollution in this regard.

This study was aimed at reviewing updated information related to the characteristics of such glycerin and its general applications, making some emphasis on its use in the soil.

PHYSICAL AND CHEMICAL CHARACTERISTICS OF GLICERIN

Pure glycerin is a carbonated-chain polyalcohol of three carbon atoms and three hydroxyl groups (CH_2OH-CHOH-CH_2OH).

Its molecule has a large number of possible reactions, due to the presence of alcoholic groups (primary and secondary) that can be replaced by other functional groups to form derivatives, such as esters, amines and aldehydes. It is stable in front of oxygen under normal atmospheric conditions, but it is converted into CO_2 and water in front of oxidants. Theoretically, it can oxidize and form eleven oxidized products of a chain with three carbon atoms. Its prolonged exposure to intense radiation in the presence of air causes its oxidation, especially in front of iron and copper ions. It has also been shown that glycerin may act as a reducing agent and hydrogen source in organic compound hydrogenation (8).

It is an odorless and colorless liquid of low environmental toxicity, soluble in water and other polar solvents, insoluble in hydrocarbons, chlorinated hydrocarbons and ethers; it is not considered a volatile liquid. It is a hygroscopic substance with neutral pH (neither hydronium cations nor hydroxyl are released when dissolved in water), which is chemically stable under normal storage and handling conditions. However, it can be explosive when in contact with oxidizing agents, such as potassium chlorate. It has a high boiling point and viscosity caused by hydrogen bonds formed between its molecules (2).

The physic-chemical characteristics of crude glycerin have been widely described by international literature. The successful application of this by-product in various spheres of economy depends on the knowledge and management of those characteristics. Their expression is very diverse. Some authors refer to them according to the type of reaction that characterizes their production (transesterification, saponification and hydrolysis) (6), whereas others refer to purification and catalysis applied, or to the nature of raw material used for biodiesel production (9). The amount of impurities present also depends on the above mentioned factors. Although transesterification process produces the lowest glycerin and the highest impurity percentages, it has the greatest application (2).

Crude glycerin is a mixture containing different amounts of glycerin (higher percentage), detergent, alcohol (mainly methanol), sodium or potassium salts (10) depending on the catalyst used, non-glycerol organic matter (NGOM) and water (11). The composition of crude glycerin records marked differences in the values (percentages or intervals) reported by different authors (3, 10, 12, 13) and the type of variables measured (Table). The beginning and end of intervals represent markedly different magnitudes for most variables.

Due to the high variability of data reported by each researcher, it is necessary to have a thorough characterization of this by-product to be used without undesirable effects. Along with the samples taken for analysis, some information is required on production process peculiarities (type of reaction used, added products, raw material characteristics for biodiesel production, among others), in order to ensure the data set is statistically representative and reliable. For instance, although the addition of glycerin improves biogas production process, the use of high ratios may adversely affect methanogenesis process, verified by microorganisms due to its high salt content (12).

Despite glycerin is a valuable by-product with more than 2000 industrial applications, it has a poor marketing due to its abundant impurities (2).

The use of crude glycerin as an energy source to obtain different chemicals through microorganisms and to synthesize chemicals, in general, are the main variants recorded by different researchers (Scheme).

BENEFICIAL EFFECTS OF APPLYING GLYCERIN TO THE SOIL

The study of glycerin as a carbon source of nitrogen fixation to the soil dates back over 50 years. A study performed on the decomposition of this polyalcohol by bacteria (27) found that it can be decomposed by various kinds of microorganisms, such as *Arthrobacter*, *Pseudomonas*, *Bacillus*, *Escherichia*, *Paracolobactrum*, *Aerobacter*, *Serratia*, *Sarcina*, *Streptomyces* and *Azotobacter*. Almost all species of *Streptomyces* and some yeasts had this same capability. The greatest bacterial growth was observed when ammonium salts were used instead of nitrate salts as a nitrogen source. In the second part of this research study (28), authors reported that certain species of *Azotobacter* fix nitrogen and decompose glycerin. This mechanism was supported by the argument that a vigorous carbon oxidation of glycerin caused a simultaneous reduction of atmospheric nitrogen.

Nitrogen-fixing microorganisms improve soil fertility and constitute a subject of study to help agricultural sustainability. The addition of glycerin to the soil may promote the survival and amount of *Rhizobium* bacteria, increase nodulation and nitrogen fixation (29).

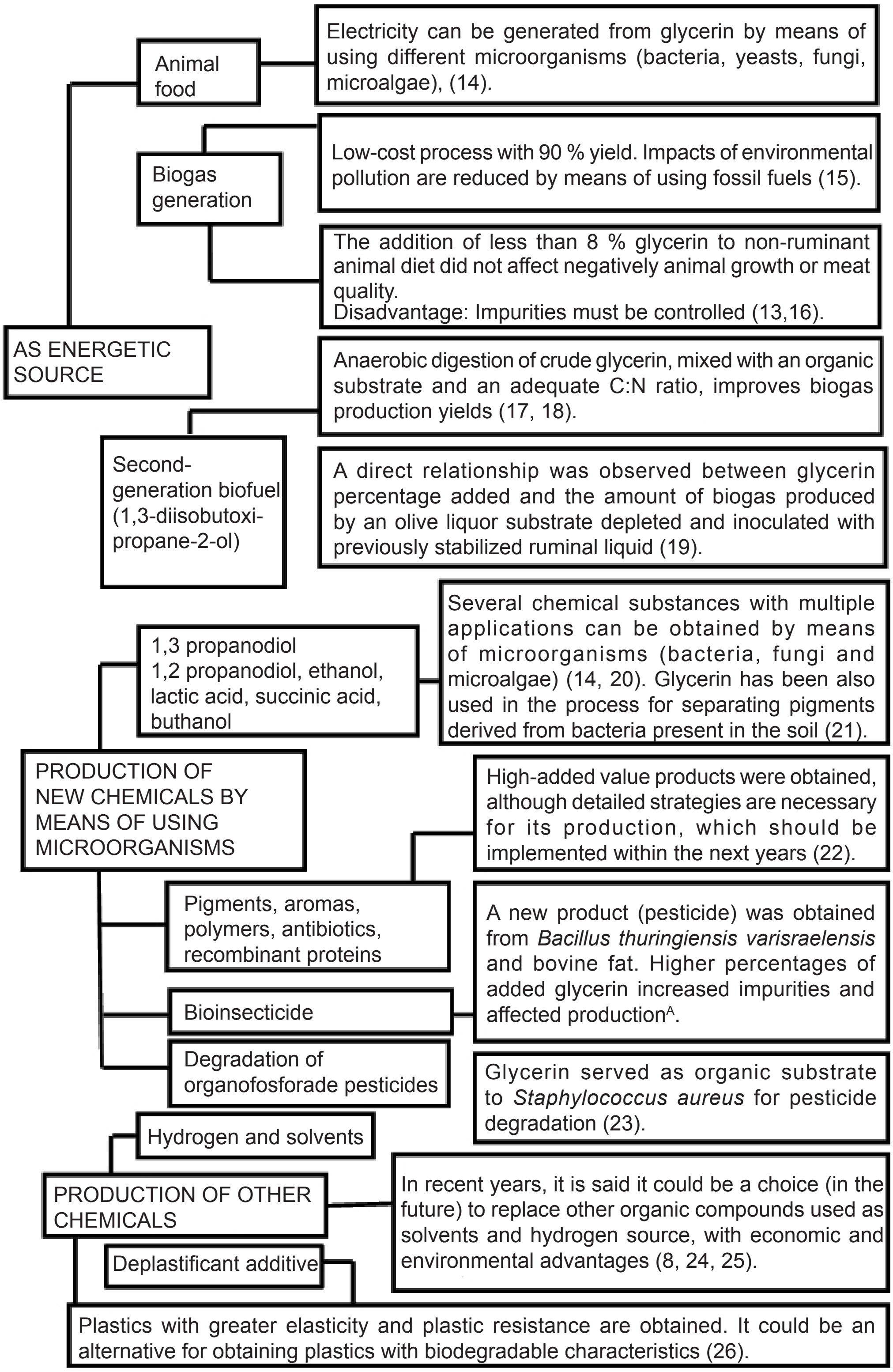
Other researchers (30) studied growth with different sources of carbon and nitrogen from a nitrogen-fixing microorganism identified as *Bacillus sphaericus* UPMB10. Results revealed that the highest yield (cell number produced per gram of carbon consumed) was achieved when glycerin was used as a carbon source. In this case, production cost was 15 times lower than when using lactates as a carbon source.

In a sandy soil treated with ammonium nitrate as nitrogen fertilizer and 1 % glycerin (percentage based on weight), it was observed that 50 % nitrogen was fixed two days after starting the study and it was completely fixed after five days (31).

Table. Crude glycerin composition according to several authors' criteria

Characteristics in percentages, according to the type of reaction used for obtaining glycerin (3, 10)				Intervals that different crude glycerin components may have (12)		Crude glycerin characteristics obtained in different biodiesel plants from Australia (expressed in %) (13)	
Component	T*	S**	H***				
Glycerin	75	83-84	88-90	K (ppm)	0-217	Glycerin	38-97
Ashes	10	8,5-9,5	0,7-1,0	P (ppm)	12-37	Ashes	0-29
Water	10	6-7	8-9	S (ppm)	14-128	Methanol	<0,01-14
NGOM	5	3-4	0,7-1,0	C (%)	24-37	pH (U)	2-10,8
Trimethyl glycol	1	0,1	0,2	N (%)	0,04-0,12	NGOM	1-57
				Protein (%)	0,05-0,44	Moisture	0-16

*Transesterification **Saponification ***Hydrolysis



[A] Rodriguez, B. C. *Avaliação do glycerol proveniente da fabricação do biodiesel como sustrato para produção de endotoxinas por Bacillus thuringiensis var Israelensis*. Tesis de Maestría, Universidad de São Pablo, São Pablo, Brasil, 2009, 131 p.

Scheme. Main applications of crude glycerin; products obtained, benefits and alternatives

On the other hand, other researchers have also found that glycerin is a suitable carbon source for microorganisms (27) and can improve soil microbial activity, with a corresponding increase in nutrient availability. This polyalcohol promotes soil retention capacity and helps nutrient adsorption. Glycerin applied to growing areas treated with potassium nitrate significantly reduces nitrate concentrations of leached waters (7). The nitrogen fixed by microorganisms at a given time can be released (31), which leads to a higher soil fertility and a lower amount of nutrients that may pollute surface and underground waters.

An experiment conducted in Murcia, Spain (32) revealed the use of glycerin enabled to increase the amounts of mineralized organic carbon and fixed or removed nitrogen by denitrification processes. In mixtures using glycerin concentrations at 4 and 6 %, there was a quick organic matter mineralization, which already occurred at 60 and 63 % respectively after 56 days. At the beginning of incubation, predominant nitrogen form was ammoniac, but nitrates predominated in all treatments (with glycerin and other carbon sources) after seven days.

In treatments with glycerin (high proportion of labile organic matter), inorganic nitrogen concentrations decreased since the first week, which suggests the appearance of nitrogen-fixing processes or denitrification. Both processes could be favored by the development of soil microorganisms.

Inorganic nitrogen losses are not only due to denitrification and nitrogen-fixing processes. The use of urea as nitrogen source may cause losses by ammonium volatilization and ammonia emission to the atmosphere. When urea is applied to the soil, there is a fast hydrolysis provoked by urease present in the soil and organic matter. Two ammonium cations are formed per each molecule of urea (33). Physic-chemical characteristics of the soil, temperature, pH and moisture can affect volatilization (34). High pH values increase volatilization. Such nitrogen loss should not be included into denitrification and nitrogen fixation, which is caused by using glycerin as a result of an increased microbial activity.

Since glycerin is an organic compound with high proportion of labile carbon, it favors the development of microorganisms, which in turn are involved and increase denitrification and nitrogen-fixing processes. Microorganisms play a vital role on the biogeochemical cycle of nutrients in the soil. Soil fertility is directly related and defined by the heterotrophic activity of microorganisms (35).

Increased organic matter content in the soil influences its physic-mechanical properties. It is considered that organic matter is the main factor affecting soil structural stability, which provides greater resistance to compaction and erosive processes as well as improves effective depth, among others[B].

Glycerin decomposition could be favored if the following aspects are considered: (i) N:C ratio, (ii) organic matter content and high soil microbial activity, (iii) glycerin proportion (it should not be high where it is applied) (36). These authors recommended an application rate of 4 m^3 ha^{-1}.

SOME ADVERSE EFFECTS OF APPLYING GLYCERIN TO THE SOIL

Crude glycerin has a methanol percentage (Table) that may be toxic to soil microorganisms and plant development.

Some studies show that glycerin additions at higher concentrations than 10 % retard growth (37). When applying 10 % glycerin, these authors observed that plant growth was similar to the experiment of soil with compost only. These results proved the need of knowing and managing crude glycerin characteristics. The table shows a wide interval between some features.

Glycerin may have large amounts of salts depending on the type of catalysis (12), which can affect agricultural sustainability if it is not properly handled. Chloride present in both sodium and potassium salts may be toxic to plants. The wide interval between pH values (from acid to alkaline) may influence denitrification and nitrogen-fixing processes (38) in a different manner and generally soil biochemistry.

[B] Balmaseda, C.; Ponce de León, D.; Martín, N. J. y Vargas, H. *Compendio de suelo*. edit. Universidad Agraria de La Habana, La Habana, Cuba, 2006, 227 p.

FINAL THOUGHTS

- ♦ The growing trend of biodiesel production has led to large volumes of crude glycerin, which represents a technological problem for its production. So far, research studies performed to find new applications are still insufficient and the amount generated is much higher than the one demanded. The need to obtain new products of commercial interest becomes essential to enhance marketing and avoid environmental damages, which involves the sustainability of biodiesel production.
- ♦ There is an incipient use of glycerin as a solvent and hydrogen source in hydrogenation reactions; thus, an increased number of investigations is needed. This application is economically attractive and environmentally friendly. It is expected that in the near future there is an increasing use of glycerin to produce new nanomaterials.
- ♦ Biotechnology represents an alternative for obtaining new products with high added value, but the biochemical aspects of processes must be deeply studied, besides drawing detailed strategies to use this by-product.
- ♦ Crude glycerin is an excellent source of calories to non-ruminant animal diet; however, its use requires an adequate control and management of the amount of salts and methanol present.
- ♦ Anaerobic co-digestion with different organic wastes can be an integrated solution to manage wastes and produce energy. Glycerin carbon increases C:N ratio and improves biogas production, but the amount of salts present should be considered, because they can inhibit methanogenesis process. The solid waste obtained from this process is rich in organic matter and deserves a study of its effect on soil fertility.

Enough research studies have been conducted in recent years aimed to evaluate the effect of adding glycerin to the soil. These studies have been mostly focused on appraising the effect of this by-product upon the increased organic matter concentrations, nitrogen fixation in the soil and its corresponding higher fertility. Nevertheless, whenever such kind of investigation is carried out, the glycerin used must be characterized. A monitoring design of this by-product is needed to ensure sample representativeness and the suitable selection of variables to be measured.

For a comprehensive assessment of glycerin effects on the soil, it is necessary to search for some aspects, such as:

- ♦ The effect of repeated application of glycerin in the same area and its possible leaching in runoff waters.
- ♦ The change of salt content in the soil as a result of its application.
- ♦ The effect of glycerin on the required nutrients for plants, such as phosphorus and others.
- ♦ Nitrogen proportion linked to denitrification, volatilization and fixation processes to the soil and its effect to mitigate environmental impacts.
- ♦ The effect of methanol concentrations (impurity present in crude glycerin) on soil biotic and abiotic factors (physical characteristics).
- ♦ Determination of other crude glycerin residual components.

At present, there are a lot of questions related to the use of crude glycerin. However, it represents a source of available carbon at relatively low cost and potentially suitable for multiple applications, which can provide economic, social and environmental benefits.

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