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Presentation of the Content

In the first article we present, *Proposal for a Circular Economy of glycerin as a by-product of biodiesel production* by TORRES-RIVERO, Ligia Adelayda, NIEVES-RIOS, Raquel and ARROYO-RODRIGUEZ José F., with adscription in the Instituto Tecnológico de Cancún, as the following article we present, *Ovum Pick Up (OPU) and in vitro embryo production in pregnant cows (Bos Indicus) at the Bachigualatito rancho, la Trinitaria, Chiapas* by NOGUEZ-ESTRADA, Juan, CORNEJO-CERVANTES Judith, VARGAS-MONTER, Jorge and SEBASTIAN-MEDINA Octavio, with adscription in the Universidad Politécnica de Francisco I Madero, as the following article we present, *Yahualica tree seedling chile production (Capsicum annuum) on different substrates and its behavior during the development of the crop* by ZEPEDA-ARIAS, José de Jesús, ARELLANO-RODRÍGUEZ, Luis Javier, RODRÍGUEZ-GUZMÁN, Eduardo and PADILLA-GARCIA, José Miguel, with adscription in the Universidad de Guadalajara, as the last article we present, *Use of Agroindustrial Waste of Orange to Obtain: Bioalcohol, Essential Oils and Activated Carbon* by BALDERAS, Elvia, GUEVARA, Elsa, LÓPEZ, Lucia and MONTOYA, Karina, with adscription in the Universidad Tecnológica Cadereyta.

Content

Article **Journal-Agrarian and Natural Resource Economics**
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Proposal for a Circular Economy of glycerin as a by-product of biodiesel production

Propuesta de una Economía Circular de la glicerina como subproducto de la producción de biodiesel

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Abstract

The Circular Economy of glycerin a proposal to reduce the environmental impact, the economic contribution, as well as avoid the discharge of used edible oils into bodies of water, the problems towards gray water treatment plants in the pretreatment process, in the landfill to prevent runoff from contaminating the soil, due to the type of Karstic soil in the region. Obtaining glycerin is one of the objectives of the project, part of the work is its application of fuel cells, as an alternative source of renewable energy. The proposed methodology Circular Economy of glycerin by-product obtained from biodiesel and the uses derived from it, the economic contribution and sustainability of this. As a result, obtaining an environmental, economic benefit, which requires both compliance with environmental laws and regulations, letting the population of the northern area of Quintana Roo know that there are alternative places to the pipes, throwing it in garbage bags where they deposit their used oil, the use of glycerin as a component livestock feed supplement as soil fertilizers and use in the generation of biogas.

Circular Economy, Glycerin, Biodiesel

Resumen

La Economía Circular de la glicerina una propuesta para reducir el impacto ambiental, el aporte económico, así mismo evitar el vertido de los aceites comestibles usados hacia los cuerpos de agua, los problemas hacia las plantas de tratamientos de aguas grises en el proceso del pretratamiento, en el relleno sanitario evitar que las escorrentías contaminen los suelos, debido al tipo de suelo Kárstico de la región. La obtención de la glicerina es uno de los objetivos del proyecto, parte del trabajo su aplicación de celda de combustible, como una fuente alterna de energía renovable. La metodología propuesta Economía Circular de la glicerina subproducto obtenido del biodiesel y los usos que de ella derivan, el aporte económico y sustentabilidad de este. Como resultado obtener un beneficio de tipo ambiental, económico, la cual requiere tanto el cumplimiento de leyes y normas ambientales, hacer saber a la población de la zona Norte de Quintana Roo que existen lugares alternativos a las cañerías, de tirarlo en bolsas a la basura donde depositan su aceite usado, él uso de la glicerina como un suplemento alimenticio de ganado componente como fertilizantes del suelo y del uso en la generación de biogás.

Economía Circular, Glicerina, Biodiesel

Citation: TORRES-RIVERO, Ligia Adelayda, NIEVES-RIOS, Raquel and ARROYO-RODRIGUEZ José F. Proposal for a Circular Economy of glycerin as a by-product of biodiesel production. Journal-Agrarian and Natural Resource Economics. 2023. 7-12: 1-5

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Introduction

Glycerin a by-product of transesterification in biodiesel production Glycerin or glycerol (propanetriol) in its pure state is a colorless, odorless, viscous and non-toxic liquid with a very sweet taste and has several uses. Pure glycerin is a polyol with a carbon chain of three carbon atoms and three hydroxyl groups (CH2OH-CHOH-CH2OH). See figure 1. 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 and form derivatives such as esters, amines and aldehydes. It is stable against oxygen under normal atmospheric conditions, but against strong oxidants it is converted into CO2 and water.

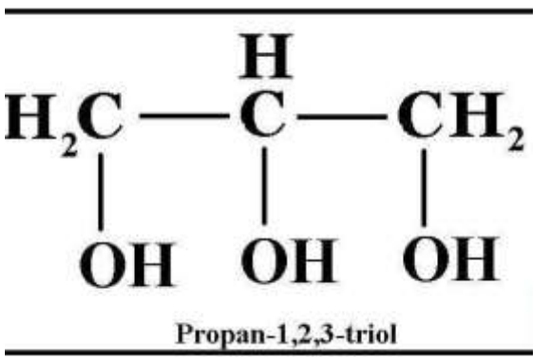

Figure 1 Glycerol propan-1, 2,3-triol - é is an organic compound belonging to the alcohol function.

Glycerin from the canola oil transesterification process is normally dark brown in color as shown in Figure No. 2 because it contains most of the substances that are not transesterifiable**.**

Figure 2 Transesterification process to obtain biodiesel and dark brown glycerin,
Own Source

Crude glycerin is a mixture containing different amounts of glycerin (higher percentage), detergent, alcohol (mainly methanol), sodium or potassium salts (Singhabhandhu, A. and Tezuka, T.2010) (Santibáñez, C.; Varnero, M. T. and Bustamante, M.2005) depending on the catalyst used, non-glycerol organic matter (MONG) and water.

The stoichiometric ratio for transesterification requires three moles of alcohol and one mole of triglyceride to give three moles of ester and one mole of glycerol; however, transesterification is an equilibrium reaction in which an excess of alcohol is necessary to drive the reaction to the product side, hence the usual conversion is 6:1, which is the first for which a complete reaction is achieved. The recovery of glycerin in the transesterification process is the fundamental part of biodiesel production, due to its industrial uses such as detergents, plastics, pharmaceuticals, studies such as Leevijit's suggest that the best results are achieved for higher ranges, between 9:1 and 12:1. In the production of biodiesel, glycerin is obtained as a by-product.

Glycerin is obtained in the transesterification process and is easily separated from the ester by decantation, requiring a purification process before its use in order to achieve the appropriate quality for its future application. See figure 3

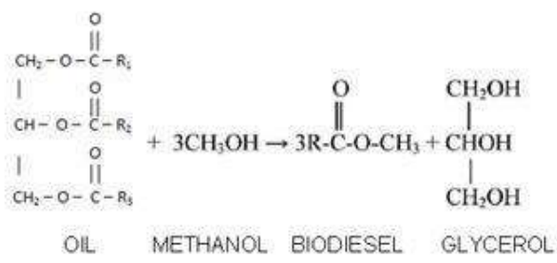

Figure 3 The transformation of vegetable oils for conversion into fatty acid methyl ester or biodiesel is carried out through the following continuous process
Source Biodis.

Used edible oils were characterized by acid number, peroxide number and dynamic viscosity, following AOAC 940.28, AOAC 965.33 and ASTM D2196, respectively. Moisture content was established by calculating the mass loss upon heating the oils (~ 5 g) from 25° to 120°C in a moisture analyzer (MX-50). González Restrepo, D. A. (2021).

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Both its purification and the search for applications of crude glycerin have surprised the scientific group, due to the product of the accelerated growth of biodiesel production and the marked tendency to increase it. The glycerin obtained from the production of biodiesel has the following characteristics a maximum concentration of 60% glycerin, is of no value, since it is which contains a large amount of soaps, alkaline catalyst and methanol and this compound is environmentally hazardous; in order to take advantage of it without prior treatment (Torres-Rivero, L. A., Ben-Youssef, B. C., & Pérez-Gasca, M. 2019).

Alternative uses are being developed for this abundant biomaterial obtained from the processing of biodiesel by acid and alkaline catalysis. Once these technologies are commercialized, the potential for improving the economics of biodiesel production, from crude glycerin, would be increased to the analysis of the nutritional value used in livestock feed.

Uses of Glycerin by-product of biodiesel.

The uses are applied in since, if it is implemented in different sectors of the economy, such as in the use of fertilizers, liquid soap to clean bathroom floors, H2 production from crude or purified glycerol.

It increases the yield of biogas production with the addition of glycerin. The applications of this by-product are the production of handmade soaps, degreaser, it is a non-irritating, biodegradable and recyclable compound.

Description of the method

The generation of large quantities of used edible oils by the restaurant industry, economic kitchens, and fast food, puts us on alert due to the lack of knowledge of the population, the bad practices on the disposal of used cooking oils, the lack of knowledge on the reuse of used oil, and the lack of knowledge on the reuse of used oil. degreaser, floor cleaner, is a non-irritating, biodegradable and recyclable compound.

Because the health of the population is at risk and, therefore, the quality of life that has been achieved by the process of industrialization, with the circular economic system, it is intended to replace the current linear system, which is only consume, use and throw away, this has led to an acceleration in the degradation of the global ecosystem. In this system, it is proposed to "reduce, reuse and recycle, and give a calm to the planet and renew its environment.

For the collection of edible oil, approximately 15 liters of edible oil were collected in the Institute's cafeteria. approximately 15 liters of residual edible oil from the cafeteria kitchen to obtain biodiesel, the final destination of the crude glycerin, see Figure 4.

Figure 4 Samples of oil collected in the Institute's cafeteria
Own Source

The entire experimental development was carried out from esterification with acid catalyst to the esterification and transesterification process to obtain biodiesel, as well as glycerin, as shown in Figure 5 below.

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Figure 5. A) Esterification process, b) transesterification process to obtain biodiesel and the by-product glycerine
Own Source

Results

Crude glycerin is the main by-product obtained during the biodiesel production process, so its chemical composition contains residues of methanol, water, soaps and salts, see Figure 6, so it has to undergo a purification process to be used in various products.

A bibliographic review was carried out, resulting in the fact that there is not enough information on the subject of eco-design mentioned in one of the sections, in order to situate this research in the most current possible scenario and give this glycerin a useful life, and prevent used edible oils from contaminating large quantities of water through dumping, disposal, as well as the contamination of the karst type soil that predominates in the Yucatan Peninsula.

Figure 6 Glycerin obtained from the production of biodiesel in the Institute's chemistry laboratory
Own Source

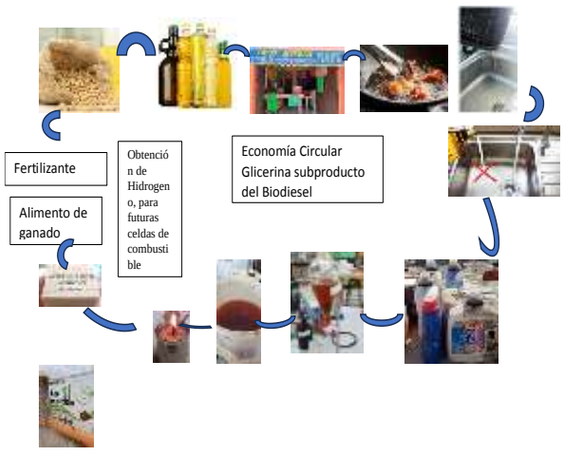

Figura 7 Below shows the circular economy proposal for glycerin as a biodiesel by-product

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Conclusions

The glycerin obtained from the biodiesel process contains residues such as methanol, salts, water, so it is of utmost importance to purify it and avoid any damage where it will be used. These pollutants, especially alcohol, are used in fuel cells to obtain H2, as a source of renewable energy. It is of utmost importance to make this proposal known to people who are interested in acquiring the glycerin obtained from the biodiesel plant and give it an added value and obtain an additional source of income, thus protecting our environment from the negative impact of waste poured into the sewers, kitchen drains and garbage without prior treatment**.** The Circular Economy proposal for the use of crude glycerin obtained from biodiesel by-products will make an economic contribution and reduce the amount of oils used in frying and reduce the disposal of the AVU, and create awareness workshops on the disposal of these oils.

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Annexes

The EC diagram is shown in Annex 1.

References

Amicarelli, V., Geatti, P. y Bux, C. (2023). El potencial de economía circular de los coproduc y subproductos de gallinas usadas en Italia mediante análisis de flujo materiales. *Ambientes* , *10* (8), 137.

Borówka, G., Semerjak, G., Krasodomski, W. y Lubowicz, J. (2023). Glicerina purificada partir de la producción de biodiésel como materia prima verde a base de biomasa o residuos pala producción de productos bioquímicos. *Energías* , *16* (13), 4889.

Broncano Morales, H. A., Cornejo Prado, P. O., Espinoza López, W. D. J., & Ríos García, P. A.(2015). Plan estratégico para la producción de biocombustibles en el Perú con enfoque de economía circular.

Cooney, R., de Sousa, DB, Fernández-Ríos, A., Mellett, S., Rowan, N., Morse, AP, ... & CliffoE. (2023). Un marco de economía circular para la valorización de residuos de pescados mariscos para enfrentar los desafíos y oportunidades para la sostenibilidad intensiva. *Revista dePproducción más Limpia*, 136283.

Hu, X., Lu, J., Liu, Y., Chen, L., Zhang, X. y Wang, H. (2023). Oxidación catalítica sostenible de glicerol: una revisión. *Cartas de química ambiental*, 1-37.

Martins, RJ y Zschach, LG (2023). Mejora de la producción de metano a partir de glicerol crudo mediante pretratamiento con ultrasonido. *Avances en la ciencia del agua*, *34* (3), 11-17.

González Restrepo, D. A. (2021). Estudio de la obtención de biodiesel como alternative energética a partir del tratamiento de residuos de aceite de cocina. Instituto Interamericano de Cooperación para la Agricultura. Informe anual 2005 = 2005 Annual Report / IICA.-. San José, C.R.: IICA, 2006. 117 p.

Labandeira Xavier, León Carmelo, Vázquez María Xose. Libro ECONOMIA MBIENTAL, editorial Pearson

Nivia-Osuna, A., Ramírez-Peña, A., Porras-Sánchez, C. J., & Marentes-Barrantes, D. L. (2020). Glicerol: suplemento alimenticio y su respuesta en bovinos de leche. *Agronomy Mesoamerican*, 821-833.

Singhabhandhu, A. y Tezuka, T. ‘‘A perspective on incorporation of glycerin purification process in biodiesel plants using waste cooking oil as feedstock’’. *Energy*, vol. 35, no. 6, junio de 2010, pp. 2493-2504, ISSN 0360-5442, DOI 10.1016/j.energy.2010.02.047.

Santibáñez, C.; Varnero, M. T. y Bustamante, M. ‘‘Glicerol Residual de la Producción de Biodiesel, Residuo o Potencial Fuente de Energía: Una Revisión’’. Chilean Journal of Agricultural Research, vol. 71, no. 3, septiembre de 2011, pp. 469-475, ISSN 0718-5839, DOI 10.4067/S0718-58392011000300019.

Article **Journal-Agrarian and Natural Resource Economics**
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Ovum Pick Up (OPU) and *in vitro embryo* production in pregnant cows (*Bos Indicus*) at the Bachigualatito rancho, la Trinitaria, Chiapas

Aspiración Folicular (OPU) y producción *in vitro* de embriones de vacas gestantes (*Bos Indicus*) en el rancho Bachigualatito, la Trinitaria, Chiapas

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Abstract

With the objective of determining the production, quality and development of bovine embryos (*Bos Indicus*), oocytes were collected from 14 females distributed in two groups: empty females (T1; n=7) and an experimental group of pregnant females (T2; n=7). The two groups were subjected to Ovum Pick Up (OPU) using disposable 18-gauge needles and a vacuum pressure of 100 mmHg. to later carry out in vitro fertilization (IVF). The study variables were the number of oocyte aspirated and fertilized and the expected embryos. A Wilcoxon test was performed for non-parametric data between two groups, finding no significant differences between treatments P> 0.05. 250 oocytes were recovered by fertilizing out of 190, obtaining a fertilization rate of 77%, reaching a transferrable blastocyst rate of 24% (45/190). In empty females (7), 113 oocytes were obtained, reaching a fertilization rate of 75% (81/113) and 25% transferable blastocysts (18/81). In the 7 pregnant females, 137 oocytes were collected, reaching a fertilization rate of 83% (109/137) and 23% transferable blastocysts (27/109). Follicular aspiration for *in vitro* fertilization of embryos from pregnant and empty cows is viable without differences between them, but being more variable in pregnant cows.

Bovine, Blastocyst, Oocytes, *In Vitro*

Resumen

Con el objetivo de determinar la producción, calidad y desarrollo de embriones bovinos (*Bos Indicus*), se recolectaron ovocitos de 14 hembras distribuidas en dos grupos: hembras vacías (T1; n=7) y hembras gestantes (T2; n=7). Los dos grupos fueron sometidos a aspiración folicular (OPU) utilizando agujas descartables de calibre 18 y una presión de vacío de 100 mmHg. para posteriormente llevar a cabo la fertilización in vitro (FIV). Las variables de estudio fueron número de ovocitos fertilizados y la previsión de embriones. Se realizó una prueba de Wilcoxon para datos no paramétricos entre dos grupos, no encontrando diferencias significativas entre tratamientos P>0.05. Se recuperaron 250 ovocitos, fertilizando 190, obteniendo una tasa de fertilización del 77%, con una tasa de blastocistos trasferibles del 24% (45/190). En hembras vacías (7), se obtuvieron 113 ovocitos, una tasa de fertilización del 75% (81/113) y 25 % de blastocistos transferibles (18/81). En las 7 hembras gestantes, se recolectaron 137 ovocitos, una tasa de fertilización del 83% (109/137) y un 23 % de blastocistos transferibles (27/109). La aspiración folicular para la fertilización *in vitro* de embriones provenientes de vacas gestantes y vacías es viable sin diferencias estadísticas entre ellas, pero con tendencias favorables con vacas gestantes.

Bovino, Blastocisto, Ovocitos, *In Vitro*

Citation: NOGUEZ-ESTRADA, Juan, CORNEJO-CERVANTES Judith, VARGAS-MONTER, Jorge and SEBASTIAN-MEDINA Octavio. Ovum Pick Up (OPU) and *in vitro* embryo production in pregnant cows (*Bos Indicus*) at the Bachigualatito rancho, la Trinitaria, Chiapas. Journal-Agrarian and Natural Resource Economics. 2023. 7-12: 6-13

† Researcher contributing as first author.

Introduction

Embryo transfer (ET) maximises herd reproduction through genetic improvement and the ability to ensure the birth of a greater number of offspring of the desired sex. In general, embryos are more resistant to heat stress than gametes, representing an advantage over artificial insemination (AI) and fixed-time artificial insemination (FTAI). ET together with the benefits of sexed semen application and *in vitro* embryo production (IVPP) are some of the main strategies to improve the livestock sector. (Zangirolamo, y otros, 2018).

IVPP and applied reproductive technologies in livestock have shown significant progress in recent years. The combination of IVPP with sexed semen (SS) and genomic selection (GS) is proving successful and widely used in countries all over the world (Ferré, y otros, 2020). The use of *in vitro* fertilisation (IVF) in bovine embryo production has increased globally to accelerate the selection of cows with high genetic values. The selection of embryos with high implantation potential is a critical factor in establishing gestation (Magata, 2023).

There are still unresolved aspects of IVEP that limit wider implementation of the technology, including low fertility due to the use of SS, reduced oocyte quality after *in vitro* maturation and low cryo-tolerance, which reduces gestation rates compared to in vivo produced embryos (Ferré, y otros, 2020).

In addition, the climatic and management conditions of the recipient females on the different farms influence the gestation rate of the transferred embryos (Pérez-Mora, Segura-Correa, & Peralta-Torres, 2020).

IVEP is a technique that makes it possible for unfertilised eggs to mature, fertilise and develop under laboratory conditions. Unfertilised oocytes and semen from selected bulls are the raw material for this technique. Under optimal conditions, about 95% of the oocytes can reach maturation, and of these, about 80% can reach the two-cell stage; however, less than half (30-50%) reach the blastocyst stage seven days after fertilisation (Ferré, y otros, 2020).

Oocyte maturation is a complex process involving nuclear and cytoplasmic modulations, during which oocytes acquire their ability to fertilise and support embryonic development. The oocyte is apparently "primed" for maturation during its development in the dominant follicle (Razza, y otros, 2019).

For best results, the technical components of transfer must be improved: nutrient quality and composition, synchronisation protocols, freezing and handling during embryo transfer. The conscious minimisation of these factors has important economic implications, favouring the efficiency of the transfer TE (Wieczorkiewicz, Jaśkowski, Wichtowska, Olszewska Tomczyk, & Jaśkowski, 2021).

The causes of low fertility in dairy cattle are complex and multifactorial, and may be due to compromised follicular development affecting oocyte quality, a sub-optimal reproductive tract environment unable to support normal embryo development, or a combination of both. (Lonergan & Sánchez, 2020).

The size of the CL influences pregnancy rate, with a higher likelihood being observed when transferring the embryo into the horn ipsilateral to a CL3 and 2, and less likely when transferring into a recipient with a CL1 < 15 mm in diameter. In addition, there is a greater likelihood of achieving pregnancy by transferring embryos at the BX expanded blastocyst and BL blastocyst stage of development, compared to BI early blastocyst and hatching developments BN (Valencia Ocampo, Rodríguez Colorado, & Mantilla, 2023).

Progesterone (P4) plays a key role in reproductive events associated with the establishment and maintenance of pregnancy through its effects on oocyte quality and its action on the uterine endometrium. Reduced P4 concentrations during ovulatory follicle growth are associated with lower fertility, and low circulating P4 concentrations after ovulation have been associated with lower pregnancy rates in cattle (Lonergan & Sánchez, 2020).

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In determining the effect of plasma progesterone (P4) on oocyte retrieval, oocyte quality and early *in vitro* embryo development in *Bos indicus* cows, the presence of higher P4 has a positive effect on oocyte retrieval, oocyte quality and early *in vitro* embryo production (IVEP) outcomes (Saad , y otros, 2019).

The efficiency of producing embryos using *in vitro* technologies in livestock species rarely exceeds the 30% to 40% threshold, indicating that the proportion of oocytes that fail to develop after fertilisation and *in vitro* culture is considerably large. The presence of cycle-related structures in the ovaries, follicle size between 6 and 10 mm, large number of cumulus cells, large oocyte diameter (>120 microns), quality of cytoplasm, structure of the perivitelline space, zona pellucida and polar corpuscle morphology have been associated with better quality. Sorting and selection of oocytes in livestock species for *in vitro* embryo production and micromanipulation techniques may be one of the most important steps in achieving superior embryo development and quality (Aguila, y otros, 2020).

In Holstein cattle, the combination of low oocyte recovery, young donor age and milk production status negatively influences IVEP. Oocyte quality is a key factor in obtaining a live calf from an in vitro embryo. Follicular wave synchronisation and the use of follicle stimulating hormone (FSH), improve oocyte quality and thus embryo production. Quality control in the laboratory and the use of high quality inputs are essential to reduce variability in production (Demetrio, y otros, 2020).

Embryo production consists of three stages, in vitro maturation (IVM) of oocytes, in vitro fertilisation (IVF) and in vitro culture (IVC) of potential zygotes, seeking to obtain quality blastocysts for greater reproductive efficiency (Salgado Cruz & Lopera Vásquez, 2020). A topic of discussion over the years is the viability of oocytes recovered by means of the Ovum Pick Up (OPU) technique from pregnant *Bos indicus* females versus oocytes obtained from empty females, because it is believed that the stimulation of progesterone produced by the corpus luteum increases oocyte viability, however, at the field level no difference has been observed in terms of in-vitro production (IVP).

Due to the limited information reported in Mexico on this subject, it is not possible to determine whether progesterone actually influences the improvement of oocyte quality and thus to have a better picture of embryo production. It is of utmost importance for the farmer to be aware of the advantages and disadvantages of subjecting pregnant females to follicular aspiration in order to be able to make decisions at crucial moments, in addition to the fact that this research can support a more accepted theory on this subject in Mexico. Therefore, in the present study the fertilisation and production of *in vitro* embryos obtained by follicular aspiration of pregnant cows (*Bos indicus*) in the Bachigualatito ranch, La Trinitaria, Chiapas, was evaluated.

Methodology to be developed

Location of the study area

The study was carried out at the Bachigualatito S.P.R. de R.L. ranch, located in La Trinitaria, Chiapas; with an altitude of 1540 m.a.s.l. (Figure 1). Fourteen *Bos Indicus* heifers were used in random stages of their estrous cycle with a Body Condition (CC) of 3 to 3.5 in a scale from 1 to 5, kept in grazing and with mineral salts supply, which were randomly distributed in two treatments: T1 empty cows and T2 Pregnant cows. Embryo production in the state of Chiapas, Mexico.

Figure 1 Batch of donor females (*Bos indicus*)

Follicular aspiration

The follicular aspiration process (Figure 2) started with epidural anaesthesia, 5 ml of 2% Lidocaine to minimise peristaltic movements and to have a better manipulation of the ovary. The perineal region was washed and disinfected and by means of a Mindray ultrasound equipment with a 5 mHz microconvex transducer which was adapted to a guide that was introduced into the vagina up to the fornix locating the ovaries via the rectum.

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With the help of the mandrel the aspiration was performed using 18 gauge needles that goes from a system to a falcon tube (corning 50 ml.) with 10 ml. of commercial flushing medium. A vacuum pump with a foot pedal was used throughout the process, applying a suction of 100 mmHg.

The aspirated cumulus-oocyte complexes (COCs) were between 2 and 10 mm in diameter.

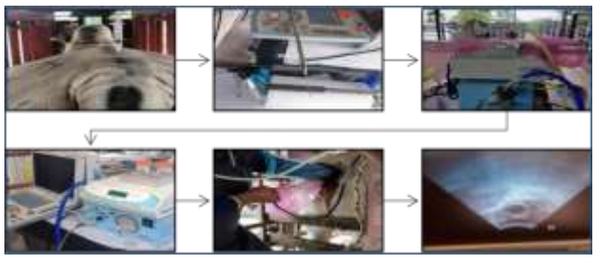

Figure 2 Methodology for follicular aspiration
Own Source

Selection and sorting

For the search, selection and sorting process, the laboratory equipment was placed in a contamination-free, sanitised area (Figure 3). All materials (pipettes, Petri dishes, falcon filter, serological pipette, 20 ml syringe) were placed on a heat platen at 37°C. At the end of oocyte aspiration, two washes were performed, tilting the collection tube at 45°, using a 20 ml flushing syringe and using a Falcon filter, placing the oocytes with the help of a serological pipette in a 30 x 40 ml Petri dish for searching with an EtScope stereoscope at 100X magnification, with a 10 µl pipette. COCs were selected and classified according to their morphology according to String fellow and Givens (2010):

Grade I are oocytes with more than three layers of compact cumulus cells and homogeneous, uniformly granular cytoplasm where the ooplasm filled the interior of the zona pellucida. Grade II are oocytes with less than three layers of cumulus cells and generally homogeneous cytoplasm. Grade III are oocytes with a single layer of cumulus cells and irregular looking cytoplasm with dark areas, the ooplasm is contracted, with space between the cell membrane and zona pellucida, irregularly filling the perivitelline space. No Cumulus bare oocytes, cytoplasm with abnormal colour and granulation and apoptotic cells, expanded are oocytes with expanded, degenerated cumulus that started their hatching process.

Oocytes were transported in 12 x 16 test tubes with medium in a TREO incubator at a temperature of 37.8 ° C.

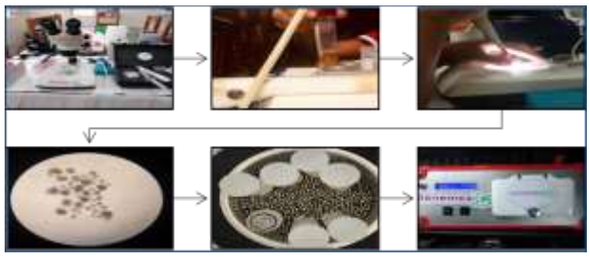

Figure 3 Oocyte search, selection and classification
Own Source

Oocyte maturation

For the in-vitro maturation process (IVM), only Grade I, Grade II and Grade III oocytes were selected, those with granular, homogeneous cytoplasm, without dark spots or clear spaces. The selected oocytes were transferred to culture plates containing HTF maturation medium, supplemented with 10% foetal bovine serum, oestradiol and follicle stimulating hormone (FSH), covering each cell with mineral oil, then placed in a NUAIRE incubator for 24 hours to be equilibrated in an atmosphere with 5% CO2, 5% O2 and 90% N2, at a temperature of 39ºC and maximum humidity (Figure 4). The theory of maturation is that the oocyte completes meiosis in response to the ovulatory LH surge. *In vitro* maturation requires 24 hours for the oocyte to complete nuclear maturation and reach the MII stage. It is also very important that there is cytoplasmic maturation, as this prepares the oocyte to support fertilisation and provide the nutrients required for embryo development.

In vitro fertilisation

Once the oocytes were matured for 24 h, conventional sperm capacitation was performed. Motile spermatozoa were obtained by centrifugation through a discontinuous gradient of Percoll 350 µl by adding 5 µl of the prepared semen in a 1 ml microcentrifuge tube at 3000 rpm for 5 minutes (Figure 4), to subsequently remove the seminal plasma components, cryoprotectants and dead or low vitality spermatozoa. The most motile fraction is enabled to penetrate the zona pellucida of the oocyte. Semen was added to each of the fertilisation cells with a pipette.

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Fertilisation plates and media were prepared prior to the start of semen capacitation. Approximately 1 million sperm per ml of media were added to the fertilisation plates (Figure 4) within the first minute after capacitation and placed in the incubator at 39ºC. Equilibrated to 5% CO2, 5% O2 and 90% N2, at maximum humidity, and cultured for a period of 20 hr.

Figure 4 *In vitro* fertilisation
Own Source

The embryo cultures were removed from the fertilisation plates and washed in new Syntetic Oviductal Fluid (SOF) medium, evaluated and the fertilised oocytes were transferred to the culture plates and placed in the incubator at 38.8ºC, equilibrated with 5% CO2, 5% O2 and 90% N2, at 100% relative humidity. On the fourth day, an evaluation of the embryos was carried out with the stereo microscope, supplementing the medium with pyruvate.

The number of blastocysts obtained was assessed at 7 days after IVF (Figure 5).

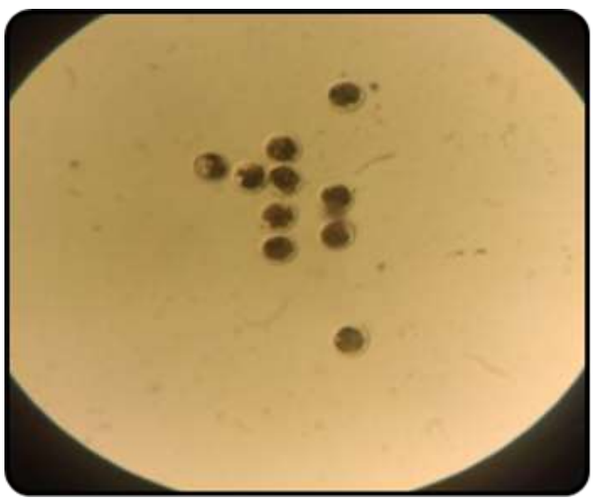

Figure 5 Blastocyst evaluation (in italics)

On day 7 of IVF, embryo transfer (Figure 6) was performed in the pre-implantation stage in the uterus of 45 recipient females.

Rectal palpation and ultrasound evaluation was performed to locate the ovary and state of the CL (corpus luteum), being in the horn ipsilateral to the C.L. where the embryo was transferred, in the lower third of the uterine horn. A TE gun was used, taking the straw by the end with a cotton seal, with the A.I. technique.

The TE gun was fitted with a sterile plastic sleeve to avoid introducing foreign bodies and microorganisms into the cow's reproductive tract. Epidural anaesthesia (5ml Lidocaine 2%) was applied between the sacral joints and the first coccygeal vertebra to block peristaltic movements and the anal sphincter muscle.

Figure 6 Methodology for embryo transfer
Own Source

Variables to be measured

The variables Oocytes aspirated, fertilised oocytes and transferable blastocysts were evaluated.

Statistical analysis

A completely randomised design was used, with the experimental unit being 14 cows.

$$Y_{ij} = \mu + Ti \quad + \varepsilon ij$$

Data were analysed by analysis of variance and comparison of means with Tukey's test ($\alpha \leq 0.05$) using SAS statistical software version 9.0. Data for all variables were analysed with the statistical package R Development Core Team (2019). R: A language and environment for statistical computing. A Wilcoxon test was performed for non-parametric data between two groups.

Results

250 oocytes were retrieved, fertilising 190 and obtaining a fertilisation rate of 77%, achieving a transferable blastocyst rate of 24% (45/190) (Table 1), of which 113 oocytes were retrieved from 7 empty females achieving a fertilisation rate of 75% (81/113) and a 25% transferable blastocyst rate (18/81).

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From the 7 pregnant females 137 oocytes were obtained achieving a fertilisation rate of 83% (109/137) and 23% transferable blastocysts (27/109). Similar fertilisation values of 733% and an embryo development rate of 35.1% were found in *in vitro* studies of oocytes collected at slaughterhouses (Fernández, Díaz, & Muñoz, 2007). In Fleckvieh heifers under tropical conditions, a blastocyst production rate between 41 and 58% is reported, with different aspiration frequencies in tropical climates (Solís-Corrales, Reinaldo, Morales, Ferrante, & Denis García, 2020).

Number	Donor Identification	Oocyte aspirate	Fertilised Oocytes	% Fertilised	Embryo forecasting	% Donor production
1	004/01	12	9	75	3	33
2	372	13	10	77	3	30
3	647	3	3	100	2	67
4	19	26	13	50	3	23
5	393	7	4	57	1	25
6	273	27	20	74	2	10
7	20	25	22	88	4	18
8*	639	15	14	93	4	29
9*	323	35	30	86	7	23
10*	509	16	15	99	5	33
11*	272	20	13	65	2	15
12*	267	35	27	77	5	19
13*	618/8	6	5	83	3	60
14*	1057	10	5	50	1	20
Total		**250**	**190**	**77**	**45**	24

Table 1 *In vitro* embryo production. Rancho Bachigualatito S.P.R. de R.L. *Pregnant females

Figure 7 shows the total mean number of fertilized oocytes for each treatment, being in cows with an average of 11.57 fertilized oocytes while, in pregnant cows, the average was 15.57 oocytes without significant differences (P>0.05), observing a greater dispersion of the data in pregnant cows.

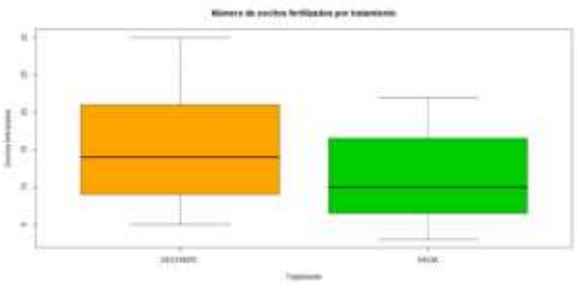

Figure 7 Number of fertilised oocytes per treatment

Figure 8 shows the total mean number of transferable blastocysts per treatment. In empty cows an average of 2.57 transferable blastocysts were obtained while in pregnant cows the average was 3.85 blastocysts.

Similar studies report 3.5 viable oocytes and 1.1 blastocysts per cow (Quispe E., Ancco G., Solano A., Unchupaico P., & Mellisho S., 2018). In OPU collection, an average of 4.3 viable oocytes is reported in Bos Taurus (Anchordoquy, y otros, 2013). In general, oocytes recovered in vivo after OPU develop better to the blastocyst stage than those obtained from ovaries in traces (Karadjole, y otros, 2010).

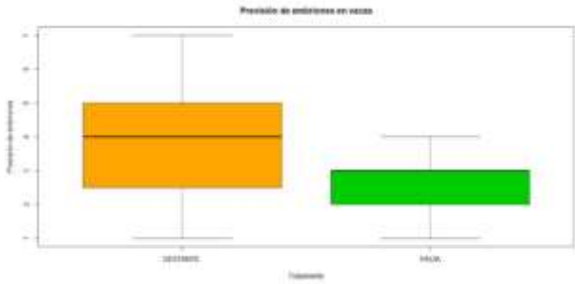

Figure 8 Number of transferable blastocysts

In studies of aspiration, procurement, selection and maturation of oocytes in vitro, it is reported that only 40% reach the blastocyst stage and of these, between 5 and 20% will produce pregnancies (Mayes M. & Sirard M., 2001). In the production of embryos, the aim is to make *in vitro* fertilisation more efficient and to mature as many oocytes as possible from in vivo aspiration in order to make the best use of donors with high genetic value without affecting their pregnancy status.

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Conclusions

Follicular aspiration for *in vitro* fertilisation of embryos from pregnant and empty cows is feasible without differences between them, however, the variability indicates that there are more factors that may be interfering and that need to be controlled and studied. The results are based on the methodology and experience to improve the embryo production programme in Zebu breeds.

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References

Aguila, L., Treulen, F., Therrien, J., Felmer , R., Valdivia , M., & Smith , L. C. (2020). Oocyte Selection for *In Vitro* Embryo Production in Bovine Species: Noninvasive Approaches for New Challenges of Oocyte Competence. *Animals., 10*(12, 2196). doi:10.3390/ani10122196

Anchordoquy, J., Huter , S., Farnetano, N., Irouleguy, J., Martín, E., Arzeno, M., . . . Munar, C. (2013). Producción de embriones por fecundación in vitro de donantes problema de razas taurinas. *Spermova, 3*(2), 126 - 129. Recuperado el 09 de 08 de 2013, de http://spermova.pe/site/files/revista2013-vol3%20No.2/126-129ok.pdf

Demetrio, D., Benedetti, E., Demetrio, C., Fonseca, J., Oliveira, M., Magalhaes, A., & dos Santos, R. (2020). How can we improve embryo production and pregnancy outcomes of Holstein embryos produced in vitro? (12 years of practical results at a California dairy farm). *Anim Reprod, 17*(3), 1-13. doi:10.1590/1984-3143-AR2020-0053

Fernández, A., Díaz, T., & Muñoz, G. (2007). Producción In vitro de Embriones Bovinos. *Revista de la Facultad de Ciencias Veterinarias, 48*(1), 51-60. Recuperado el 08 de 08 de 2023, de http://ve.scielo.org/scielo.php?script=sci_arttext&pid=S0258-65762007000100006&lng=es&tlng=es.

Ferré, L. B., Kjelland, M. E., Strøbech, L. B., Hyttel , P., Mermillod, P., & Ross , P. (2020). Review: Recent advances in bovine in vitro embryo production: reproductive biotechnology history and methods. *Sciencedirect, 14*(5), 991-1004. doi:10.1017/S1751731119002775.

Karadjole, M., Getz, I., Samardžija, M., Maćešić, N., MatkovićMatković, M., Makek, Z., . . . Poletto, M. (2010). The developmental competence of bovine immature oocytes and quality of embryos derived from slaughterhouse ovaries or live quality of embryos derived from slaughterhouse ovaries or live donors by ovum pick up. *VETERINARSKI ARHIV, 80*(4), 445-454. Recuperado el 09 de 08 de 2023, de chrome-extension://efaidnbmnnnibpcajpcglclefindmkaj/https://hrcak.srce.hr/file/88435

Lonergan, P., & Sánchez, J. M. (2020). Symposium review: Progesterone effects on early embryo development in cattle. *J. Dairy Sci., 103*(9), 8698-8707. doi:10.3168/jds.2020-18583

Magata, F. (2023). Time-lapse monitoring technologies for the selection of bovine in vitro fertilized embryos with high implantation potential,. *Journal of Reproduction and Development, 69*(2), 57-64. doi:10.1262/jrd.2022-131

Mayes M., A., & Sirard M., A. (01 de 03 de 2001). The influence of cumulus-oocyte complex morphology and meiotic inhibitors on the kinetics of nuclear maturation in cattle. *Theriogenology, 55*(4), 911-922. doi:10.1016/s0093-691x(01)00453-8

Pérez-Mora, A., Segura-Correa, J. C., & Peralta-Torres, J. A. (2020). Factors associated with pregnancy rate in fixed-time embryo transfer in cattle under humid-tropical conditions of México. *Anim. Reprod., 17*(2), 1-9. doi:10.1590/1984-3143-AR2020-0007

Quispe E., C., Ancco G., E., Solano A., J., Unchupaico P., I., & Mellisho S., E. (2018). Capacidad de desarrollo embrionario de ovocitos de bovino recuperados vía ultrasonografía y de ovarios de matadero. *Rev. investig. vet. Perú, 29*(4), 1114-1121. doi:http://dx.doi.org/10.15381/rivep.v29i4.14418

Razza, E. M., Pedersen, H. S., Stroebech, L., Fontes, P. K., Kadarmideen, H. N., Callesen, H., . . . Hyttel , P. (2019). Simulated physiological oocyte maturation has side effects on bovine oocytes and embryos. *J Assist Reprod Genet*(36), 413–424. doi:10.1007/s10815-018-1365-4

Saad , M., Sarwar, Z., Saleem, M., Arshad, U., Shahzad, M., Mushtaq, M. H., . . . Ahmad, N. (2019). Effect of plasma progesterone on oocyte recovery, oocyte quality, and early in-vitro developmental competence of embryos in Bos indicus dairy cows. *Animal Reproduction Science, 202*, 80-86. doi:10.1016/j.anireprosci.2019.02.001

ISSN 2524-2091

Salgado Cruz, E., & Lopera Vásquez, R. (jul-sep de 2020). Aspectos esenciales sobre las técnicas de fertilización in vitro en bovinos. *Rev. investig. vet. Perú, 31*(3). doi:10.15381/rivep.v31i3.17138

Solís-Corrales, A., Reinaldo, D., Morales, J., Ferrante, M., & Denis García, R. (2020). Producción in vitro de embriones en novillas felckvieh bajo condiciones tropicales. *Revista investigaciones pecuarias, 3*(1). Recuperado el 08 de 08 de 2023, de http://portal.amelica.org/ameli/jatsRepo/222/2221956004/index.html

Valencia Ocampo, H., Rodríguez Colorado, N., & Mantilla, T. (2023). Factores que afectan la tasa de preñez mediante transferencias de embriones por fertilización in vitro en novillas multirraciales en condiciones de trópico colombiano. *Rev Mex Cienc Pecu, 14*(2), 326-338. doi:10.22319/rmcp.v14i2.6031

Wieczorkiewicz, M., Jaśkowski, J., Wichtowska, A., Olszewska Tomczyk, M., & Jaśkowski, B. (2021). Effectiveness of embryo transfer in cows - risk factors including in vivo derived and in vitro produced embryos. *Medical Journal of Cell Biology, 9*(3), 123-131. doi:10.2478/acb-2021-0017

Zangirolamo, A., Souza, A., Bergamo, L., Silva, N., Sánchez , T., & Seneda, M. (2018). Applications of embryo transfer biotechnology in dairy cattle. *Spermova, 8*(1), 29-32. doi:10.18548/aspe/0006.03

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Yahualica tree seedling chile production (*Capsicum annuum*) on different substrates and its behavior during the development of the crop

Producción de plántula de chile de arbol Yahualica (*Capsicum annuum*) en diferentes sustratos y su comportamiento durante el desarrollo del cultivo

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Abstract

In order to observe the differences between substrates in the production of Yahualica chili pepper (Capsicum annuum) seedling and plant, in the field and greenhouse at the University Center of Cs. Biological and Agricultural of the University of Guadalajara, this investigation was carried out, where six substrates were evaluated: Promix-GLX, Promix-FLX, Sunshine-3, Berger-BN2, Germinaza and 50% Germinaza + 50% Earthworm Humus. In seedlings, the following were evaluated: Emergence Speed, Emergence Percentage, Stem Thickness, Seedling Length, Number of Leaves, Fresh Weight Aerial Part, Fresh Weight root, aerial part dry weight and root dry weight, and during plant development in field and greenhouse: Plant height, and Fresh weight of ripe fruit. Significant differences were found between treatments (P≤0.05) in the seedling and plant quality variables. Consequently with the mixture of 50% Germinaza + 50% Earthworm Humus, Berger BN2 and Sunshine 3, the best seedling quality was obtained. And moreover the best plant development was observed in the greenhouse, with an average plant height of 139 cm and a fresh fruit yield of 651 g. When germinaza was combined with earthworm humus (50:50), the results were significant to obtain vigorous plants, high percentage of emergence.

Substrates, Emergence, Agricultural, Seedling quality, Humus, Significant, *Capsicum annuum*

Resumen

Con el objetivo de observar las diferencias entre sustratos en la producción de plántula y planta de chile Yahualica (*Capsicum annuum*), en campo e invernadero del Centro Universitario de Cs. Biológicas y Agropecuarias de la Universidad de Guadalajara se llevó a cabo esta investigación, donde se evaluaron seis sustratos: Promix-GLX, Promix-FLX, Sunshine-3, Berger-BN2, (Germinaza) y 50% Germinaza + 50% Humus de Lombriz. En plántula se evaluó: Velocidad de emergencia, Porcentaje de Emergencia, Grosor de tallo, Longitud de plántula, Numero de hojas, Peso fresco parte aérea, Peso fresco de raíz, Peso seco parte aérea y Peso seco de raíz, y durante desarrollo de planta en campo e invernadero, las variables fueron: Altura de planta, y Peso fresco de fruto maduro. Se encontraron diferencias significativas entre tratamientos (P≤0.05) en las variables de calidad de plántula y de planta. La mezcla de 50% Germinaza+50% Humus de lombriz, Berger BN2 y Sunshine-3 se obtuvo la mejor calidad de plántula. El mejor desarrollo de planta se observó en invernadero, con altura de planta promedio de 139 cm y un rendimiento de fruto fresco de 651 gr. En el tratamiento de germinaza cuando se combinó con humus de lombriz (50:50), los resultaron fueron significativos para obtener plántulas vigorosas y alto porcentaje de emergencia.

Sustrato, Emergencia, Agricultura, Calidad de plántula, Humus, Significancia, *Capsicum annuum*

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Introduction

The increase of the world population forces mainly the agricultural sector to generate new technologies in order to increase the horticultural yield per unit area and the quality of food products for the demanding market. According to Guimarães et al. (2002); Hartmann and Kester (2002), in the production of high quality vegetables, seedling production is one of the most important stages in the crop cycle, since it has a significant influence on the responses of the plant, from a nutritional and productive point of view, given that there is a direct relationship between healthy and vigorous seedlings and production in the field.

In this respect, Abad and Noguera (2000) point out that soil is the natural medium for seedling growth, due to various factors such as: availability, cost, ease of obtaining, etc. However, it is not the most suitable material for seedling growth. However, it is not the most suitable material for the production of seedlings in greenhouses. From an environmental point of view, the most important criteria for choosing a material as a substrate for soilless cultivation are: its durability and its capacity for subsequent recycling. One of the most important functions of a substrate in a soilless cultivation system is to provide an "ideal" environment for the plant for root growth and to constitute a suitable base for the anchorage or mechanical support of the plants.

The selection of the substrate is one of the most important factors in providing the appropriate conditions for root growth (Ocampo et al., 2005). In agriculture, a wide variety of substrates are used for seedling production, some of the best known being: rice husks, tree bark, coffee pulp, coconut fibre, peat, sawdust and shavings, sand, gravel, earthworm humus, leaf litter, manure, among others (Acosta et al., 2008). Finding an ideal substrate is a difficult task, because each species has different requirements, but through experiments it is possible to find an optimal substrate that meets the minimum conditions required by the species to be studied (Oliverio, 2014). According to Gomes et al. (2008) and Moreno-Resendez et al. (2008), one of the most commonly used substrates for the commercial production of vegetable seedlings is peat.

Its physical, chemical and biological characteristics allow excellent germination and seedling growth, but its high cost and unsustainable exploitation have begun to restrict its use (Fernández et al., 2006). However, in Mexico there are alternative materials with physical and chemical properties that can substitute Peat Moss, such is the case of coconut powder, whose advantages include its wide availability in Mexico and adequate physical and chemical properties (Noguera et al. 2003).

One of the main horticultural crops in Mexico is the Capsicum annuum chilli, with a production of 1.85 million tonnes (López-Baltazar et al., 2013). The development of poor quality seedlings is one of the production factors that cause the most problems for producers of Yahualica chilli de árbol in the region of the crop's denomination of origin (Yahualica, Jalisco); seedlings are generally produced in seedbeds on the ground and develop very slowly due to the poor root system. As a result, many of the plants die before they can take root in the soil once transplanted and have to be replanted, increasing production costs. Although some farmers are already producing their seedlings in trays and the problem of deficient roots has improved, there are still no studies on the best substrate available on the market to solve this problem. For this reason this work was carried out, with the aim of using the results to help growers in the selection of the best substrate for seedling production of this crop.

Objective

To evaluate the effects of six substrates on seedling production and plant productivity of Yahualica chile de árbol.

Materials and methods

The research was carried out in experimental fields and greenhouses of the CUCBA of the University of Guadalajara, in the property Las Agujas, municipality of Zapopan, Jalisco, located in the central region of the state, in the extreme coordinates of 20º25'30" to 20º57'00" north latitude and 103º19'30" to 103º39'20" west longitude, at an altitude of 1,548 metres above sea level.

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The climate of the experimental site is temperate sub-humid (A) C (w1) to (e) g according to the Köppen climate classification, with an average annual temperature of 18º to 22° C, with an average rainfall of 851 mm and four days of frost in January and February (García, 2004).

Chili fruit collections. With various producers in Yahualica, a collection of dry and ripe chilli peppers was carried out, with high health and good physical appearance. The following activities were carried out in the CUCBA seed laboratory to ensure that the seed to be used had high germination power and vigour.

Obtaining pure seed: The dried fruits were threshed and the seed was separated from the impurities using sieves and wind. The seed was sorted through a pneumatic separator to obtain seed of high specific weight.

Six substrates were tested in this research:

1. Promix GLX : Canadian Sphagnum peat (90-95%/vol) 2. Vermiculite - horticultural grade; Macronutrients; Micronutrients; Dolomitic and Calcitic lime; Wetting agent. pH range 5.0 - 6.0 (SME). Electrical conductivity 0.7 - 1.1 mmhos/cm (SME). Air porosity: 6-10% by volume. Water holding capacity: 70 - 85% by volume.
2. Promix FLX: Sphagnum peat moss, limestone for pH adjustment and wetting agent.
3. Sunshine: Plug-grade Canadian Sphagnum Moss peat, plug-grade vermiculite, dolomitic limestone.
4. Berger BN2: Selected fine-grained peat moss of excellent quality, fine-grained perlite, fine-grained vermiculite, dolomitic and calcitic limestone, non-ionic humectant, initial charge of standard fertiliser for seedlings.
5. Germinaza: Ground coconut tow
6. Germinza + earthworm humus: 50% and 50%.

The research was carried out in two stages:

A. Seedling production:

The experiment was set up twice. Using 200-cavity unicel trays. Three replicates per substrate and 100 seeds per replicate were sown under a completely randomised design.

According to Reveles (2005), the seedling is ready when it has 3 to 4 pairs of true leaves and a height between 10 and 12 centimetres, which is achieved between 40 and 50 days after sowing.
Variables measured:.

– As a test of vigour, the variable speed of emergence (VE) was measured. This was obtained by counting the number of seedlings emerged daily after sowing, taking as emerged seedlings those that protrude from the substrate. The speed of emergence was calculated using the expression proposed by Maguire (1962):

$$VE= \sum_{i=1}^{n} Ni\ /ti$$

Where:

VE = rate of emergence; N_i= Number of seedlings emerged per day; ti=Number of days after sowing; n= Number of counts 1, 2....., n counts.

– Emergence percentage (EP): total number of normal seedlings emerged at the end of the test per treatment and replicate.

– Seedling length (LP): Measured from the insertion of the hypocotyl to the tip of the apex of the last leaf.

– Stem thickness (GT): Using a TRUPER digital vernier (CALDI-6MP), the diameter of the seedling was measured in the middle part of the hypocotyl.

– Number of leaves (NH). The number of leaves developed above the cotyledons was counted.

– Fresh weight and dry weight of aerial part and root (PFRA, PFPA, PSRA and PSPA) of the seedlings produced:

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In each treatment, five subsamples of 10 seedlings per treatment were weighed to obtain both aerial and root fresh weight of the seedlings. The seedlings were carefully cleaned by separating the aerial part from the roots and weighed on an analytical balance.

For the seedling dry weight test, five subsamples of 10 seedlings per treatment (representative of each replicate) were tested. The stem was then separated from the root, and these were placed separately in small brown paper bags, then taken to the oven to dry for three days at a temperature of 70ºC. After this time they were removed from the oven, allowed to cool and weighed on an analytical balance to obtain the dry weight of each treatment of both roots and aerial part.

A. Plant production in the field and greenhouse

Under field conditions, 30 seedlings per treatment/repetition were sown in a randomised block design with three replications.

In the greenhouse, 12 plants per treatment/repetition and with three replicates were transplanted into 10 litre pots using 75% jal and 25% worm castings as substrate, under a completely randomised design.

In these two environments, the 100% Stainer solution, which was prepared from calcium nitrate, potassium nitrate, magnesium sulphate, potassium sulphate and micronutrients, was fertilised using a venturi.

Both in field and greenhouse conditions, the following variables were considered: plant height (PA) sampled in two and three phases respectively, considering the measurement from the base of insertion in the substrate to the tip of the last leaves. Total fresh fruit weight (TFFW): The sample consisted of 10 plants per treatment and repetition, considering that the plant had one hundred percent of red fruits.

Statistical analysis: An analysis of variance (ANOVA) was carried out on all the variables and, where statistical differences were found, a multiple mean test was carried out using Tukey's test at 0.05 probability. The SAS 1981 statistical package was used for this purpose.

Results and discussion

Seedling production. In research carried out by Momirovic et al. (2000), Guzmán and Sánchez (2003) and Díaz et al. (2008), the variables germination percentage, seedling length, root dry weight and aerial part dry weight are considered as important variables to evaluate the quality of seedlings in chilli. Statistical differences ($P \leq 0.01$) were observed in the variables speed of emergence (VE) and percentage of emergence (PE) evaluated during two stages. This indicates that at least one substrate showed differences with the rest of the substrates. With a mean of 83 and 82% in the variable percentage of emergence within the two evaluations. And a coefficient of variation of 3.31% and 4.93% respectively considered as very good, which gives us an idea of high reliability in the results. On the other hand, the coefficients of variation of 19.6% and 15.2% were obtained for the speed of emergence during the two evaluations, and due to the nature of this variable, they can be considered as having good reliability.

In the variables stem thickness (GT), seedling length (LON), aerial part fresh weight (PFPA), root fresh weight (PFRA), aerial part dry weight (PSPA) and root dry weight (PSRA), statistical differences were found ($P \leq 0.01$) between the substrates evaluated, only in the variable number of leaves (NH) there were no statistical differences.

Means of 2.1 g, 11.45 mm, 9.7, 6.26 g, 2.97 g, 0865 g, and 0.291g respectively were observed in the variables stem thickness, seedling length, number of leaves, aerial part fresh weight, root fresh weight, aerial part dry weight, and root dry weight. And with coefficients of variation that give reliability to the results.

Mean test:

a) Variable speed of emergence (as a measure of vigour) and percentage of emergence

In table 1 for the variable speed of emergence, it was observed that the greatest vigour was presented with the combined substrate of 50% germination and 50% earthworm humus in the two tests carried out.

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This was followed by the sunshine, Berger BN2 and germination treatments. It was observed that the substrate Promix FLX and Promix GTX had a negative effect on the speed of seedling emergence by obtaining very low vigour values (Table 1).

In the two tests for the variable percentage of emergence, the substrates 50% germination + 50% earthworm humus, Berger BN2 and germination (Table 1) stood out, with emergence percentages of over 88%, while the lowest number of emerged plants corresponded to Promix FLX and Promix GTX, with germination averages of 56 and 74%. These two variables evaluated are the basis of horticultural crop production, as seedlings of high vigour and germination are needed to ensure high yields in the field. As reported by Andrade-Rodriguez et al. (2008) when using different substrates for papaya seedling production, they found that substrates in which earthworm humus was used produced better seedling growth.

b). Variable seedling length, stem thickness and number of leaves

In the variable stem length, according to graph 1, a marked difference is observed between the six substrates evaluated, where the best substrate corresponded to 50% germination + 50% earthworm humus and Berger BN2; while the germination substrate had the smallest seedlings. These results are in contrast to García et al. (2001), who tested substrates with rice husk, coconut fibre, pine bark, peat, and garden compost combined with inorganic materials, and found higher productivity and quality of Epipremnum aureum and Spathiphyllum wallisii plants when using coconut fibre, peat and peat with agrolite.

On this point, Velasco-Velasco et al. (2001) point out that the improvement observed in the growth of seedlings by adding earthworm humus to the germination substrate suggests that it can be used as a biofertiliser, thus reducing the use of chemical fertilisers. It is also considered to improve the soil as it provides organic matter and modifies physical and chemical properties (López-Moctezuma et al., 2005).

Ortega-Martínez et al. (2010), found that the variable seedling length in tomato, the information obtained at 30 days expressed significant differences.

The seedlings with the greatest height were obtained in earthworm humus (17 cm) and peat (15 cm) followed by those grown in sawdust (12 cm). On the other hand, the treatment 50% germinza + 50% earthworm humus compared to the treatment with only germinza (where small and chlorotic seedlings were obtained) showed that the addition of earthworm humus to the latter had a significant effect on seedling growth and seedling colouring.

Andrade-Rodriguez et al. (2008), in a study of papaya seedling production, the smallest seedlings were obtained when grown on coconut fibre/sawdust/agrolite (5:2.5:2.5) and coconut fibre/sawdust (7:3), where the seedlings showed general chlorosis due to the phenols released by the coconut fibre.

In the variable stem thickness, the highest value of the seedling was observed in the Promix FLX treatment (2.51 mm); and the lowest thickness was observed in the germination substrate (Figure 1), showing a value of 1.83 mm; which when compared with the substrate where worm castings were added, an increase in stem thickness of 2.2 mm was obtained (Figure 1). In this regard, Ortega-Martínez et al. (2010) found that the substrates worm castings, peat and sawdust significantly favoured a greater development in stem thickness. As for the number of leaves, as shown in graph 2, the lowest value was found in the germination treatment. However, when worm castings were added to this treatment, there was an increase in the number of leaves (from 8.6 to 11.07).

In this regard, Velasco-Velasco et al. (2001), studied the effect of the incorporation of earthworm humus and observed that it had a positive effect on the photosynthesis rate, dry matter accumulation and yield of tomato husk. According to Larqué-Saavedra et al. (2010), Parra-Terraza et al. (2010), and Berrospe-Ochoa et al. (2012), tomato seedlings at 31 to 35 d of age presented on average between three and eight leaves. Ortega-Martínez et al. (2010). They point out that in an investigation with tomato, the number of leaves produced by the seedlings in the period analysed, had significant differences caused by the treatments under study, of which the worm humus substrate achieved the highest number of leaves (7).

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Thus, the results obtained, as well as previous research, indicate the need to use in the substrates some component that provides nutrients for plant growth, in addition to adequate support.

c). Variables Fresh weight and dry weight of the aerial part and roots

In these variables, the sunshine substrate presented the highest values in comparison with the others, followed by the substrates Promix FLX, Berger BM2, Promix GTX and worm castings 50% + germinaza 50% (Table 2). While the substrate germinaza showed the lowest values in these four variables. In this regard, Ortega-Martínez et al. (2010), in an investigation with various substrates and in tomato cultivation, the dry weight of tomato seedlings showed significant statistical differences, where the best substrates were peat and earthworm humus followed by sawdust.

Higher dry matter accumulation indicates better seedling quality for transplanting and higher rooting percentage (Herrera et al., 2008; Rosca, 2009). According to Muñoz (2002), it seems that one of the best parameters is the dry matter (DM) content, so the higher the dry matter content, the more resistant the plant is to stress. It is clear that the D.M. content has a significant influence on the rooting of the plant. This will favour the development and proper growth of the other vegetative structures of the seedlings, such as leaf area weight, number of leaves and stem thickness.

Plant production and development under greenhouse and field conditions: In the analysis of variance, statistical differences were found in all the variables studied. In terms of plant height in the greenhouse, a greater average was achieved than in the field (135 cm and 109 cm respectively), and a greater yield of fresh fruit compared to that obtained in the field (651 g to 588 g respectively). In this regard, Bowman and Paul (1983) point out that there are differences between a plant grown in the field compared to one grown in a pot, which is generally exposed to a more stressful and constantly changing environment. Where the volume of a pot is limited, the substrate and its components must possess physical and chemical characteristics which, combined with a comprehensive management programme, allow optimum growth.

Comparative test of means

a) Plant height

Greenhouse: In the greenhouse area, two samples of plant height were taken in June (beginning of flowering) and in October (harvest). In the field, three samples were taken. Two in June and one in October (harvest).

As can be seen in graph 3, the plants from the substrates 50% Germinaza + 50% earthworm humus, Sunshine and Berger BN2 obtained the greatest heights (145, 145 and 140 cm respectively). While the Germinaza and Promix GTX treatments had the lowest plant heights (123 and 121 cm respectively).

Field

During the first two sampling dates the plant height of the six treatments remained constant (very similar data), but on the third date we found a marked difference between Germinaza and Promix FLX with values of 113 and 99 cm respectively. While, Promix FLX, Berger BN2, 50% Germinaza + 50% earthworm humus, Promix GTX, and Sunshine and showed the highest and very similar values in plant height at harvest (113, 113, 112, 109, and 109 cm respectively) (Figure 4).

Comparison of greenhouse and field plant height

When comparing the heights at harvest (fresh fruit) between greenhouse and field conditions, a marked difference is observed in the values obtained (Figure 5). Here, higher values can be seen in this variable under greenhouse conditions. Three groups of significance were formed: A AB and B, where the highest value corresponds to 50% Germinaza + 50% worm humus, Sunshine and Berger BN2, grouped in significance group A, and in group B the lowest value corresponds to Promix GTX and Germinaza. Under field conditions, three significance groups were also formed: A AB and B. The Germinaza treatment was placed in group B, it was the treatment with the lowest height (Figure 5). The highest values in the field were found in group A with the substrates Promix FLX, Berger BN2, and 50% Germinaza+50% earthworm humus. These last two treatments showed the most stable behaviour in the two test environments.

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According to Muñoz (2002), the environmental conditions inside the greenhouse are relatively homogeneous and controlled, but outside in the final environment they will be very heterogeneous and not very or not at all controllable. This is demonstrated by the fact that a lower coefficient of variation was obtained in the greenhouse than in the field (4.2% and 5.2% respectively).

Fresh fruit weight: In this variable under field conditions, the best treatment corresponded to Berger BN2 (855 g), 50% Germinaza + 50% earthworm humus (840 g), and Promix GTX (614 g), and the lowest harvest value (420 g) was observed in the Sunshine substrate. While, in the greenhouse, the highest value (1163 g) was observed in the 50% germination and 50% worm castings treatment, followed by Sunshine (1011 g), and Berger BN2 (937 g). The lowest weights were observed in the plants from the Germinaza and Promix GTX substrates (Figure 6). In both environments, the 50% Germinaza+50% worm castings treatment stood out. In this regard, Domínguez et al. (2010) point out that it has been shown that the addition of earthworm humus to soils and substrates considerably increases the growth and productivity of a large number of horticultural crops.

According to graph 6, there are marked differences in fresh fruit weight in the two environments. In the greenhouse, yields were much higher than those obtained in field conditions, due to the fact that greenhouse crop production reduces the effects of extreme climates, allows the production of vegetables throughout the year and increases quality and yield (Rojas and Paniagua, 2015).

In graph 6, it is observed that the Sunshine treatment, despite showing a high weight in the greenhouse, had a significant drop when produced in the field. In this regard, Bowman and Paul (1983) point out that there are many differences in a field crop and another in greenhouses and pots. In greenhouses, temperatures and humidity are controlled, favouring yields and crop development. Nesmith and Duval (1998), point out that the variability of models associated with the use of certain substrates implies different morphological and physiological responses in the field or in the greenhouse.

A quality seedling is one that has good root development, a vigorous stem, no chlorosis, and is free of pests and diseases. To overcome transplanting stress, it must have adequate root capacity for water and nutrient uptake, as well as the ability to generate new roots. The growth rate is reduced when low quality seedlings are used (Leskovar, 2001). In this regard, Pérez et al. (2007), in a study with maize varieties, observed a positive correlation between vigour variables and grain yield and its components. The variables best correlated with vigour were speed of emergence, length and mesocotyl dry weight. In this greenhouse study, jal and worm humus were used as substrate (75% and 25%, respectively).

The addition of worm humus in the growth medium and the consequent increase of nutrients in the tissue has been observed in studies with various vegetables, attributing this increase to the presence of nutrients in the worm humus, as well as to the high cation exchange capacity it possesses, in addition to high moisture retention (Pérez-Murcia et al, 2006; Grigatti et al., 2007). Due to these properties attributed to the composted materials, the addition of 100% Steiner's nutrient solution improved plant development. For their part, Ramos et al. (2011) report that the application of worm humus, in addition to adding organic matter, which influences aeration, humidity and resistance of degrading soils, improves the capacity to regulate pH and CEC, as well as favouring the availability of phosphorus, potassium, iron and manganese in the crop.

Conclusions

Differences were found in the behaviour of the substrates with respect to Yahualica chilli seedling quality, where the highest seedling quality was achieved with the substrates 50% germination + 50% earthworm humus, Sunshine, and Berger BN2.

In the greenhouse, plant development reached the highest values for plant height and fresh fruit weight. Under greenhouse conditions, mean plant height values of 135 cm and fresh fruit weight of 652 g were obtained. In the field, mean plant height values were 109 cm. And with an average fresh fruit weight of 588 g.

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In the greenhouse, the plants produced in the substrates 50% Germinaza+50% worm castings, Sunshine and Berger BN2 stood out for their height. In field conditions, seedlings produced on the substrates Promix FLX, Berger BN2 and 50% Germinaza + 50% earthworm humus stood out.

In terms of fresh fruit yield, the plants produced from the substrates 50% Germinaza + 50% earthworm humus, Sunshine and Berger BN2 achieved the highest yields under greenhouse conditions. In the field, the substrates Berger BN2 and 50% Germinaza+50% worm castings stood out.

Annexes

Treatments	Emergency Speed (1)	Emergency Speed (2)	Emergency Percentage (1)	Emergency Percentage (2)
Promix GLX	22.61 a	22.66 c	74.00 c	75.33 c
Promix FLX	8.38 b	9.64 d	60.00 d	52.00 d
Sunshine	31.56 a	31.33 abc	86.00 b	86.00 bc
BergerBN2	31.46 a	34.80 ab	93.00 ab	93.33 ab
Germinaza	27.25 a	25.92 bc	88.33 b	88.33 ab
Germinaza 50%+Wormwormworm compost 50%.	36.75 a	39.75 a	97.67 a	97.67 a
DMS	14.18	11.41	7.56	11.01

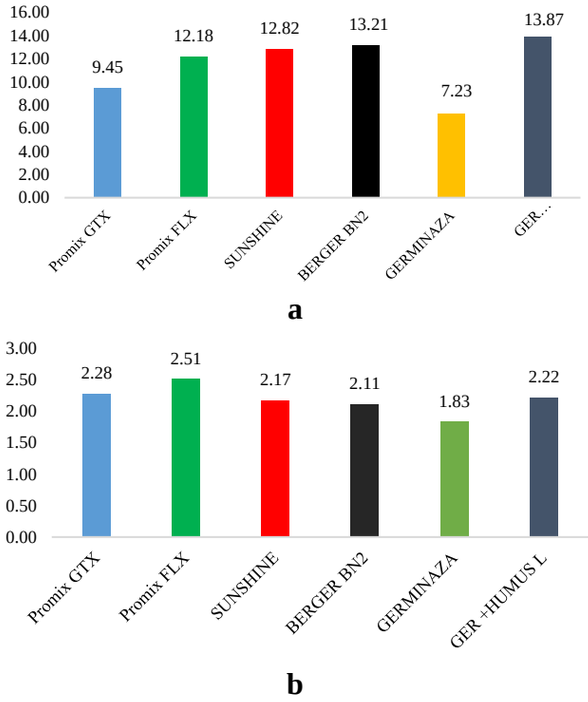
Where: Means with the same letter within columns are statistically equal, according to Tukey's test (p≤ 0.05), LSD= Least Significant Difference.

Table 1 Comparative test of means (Tukey at 0.05 probability) for the variables speed of emergence (first and second evaluation) and the variable percentage of emergence (first and second evaluation) in the six substrates evaluated

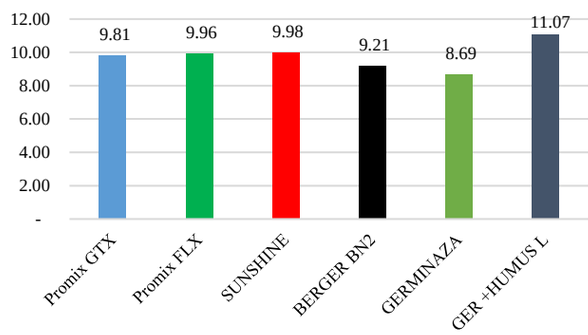
Tratamientos	PFPA	PFRA	PSPA	PSRA
Promix GLX	5.54 bc	2.36 b	0.628 c	0.212 b
Promix FLX	6.26 abc	2.74 b	0.766 bc	0.262 b
Sunshine	7.70 a	4.7 a	1.130 a	0.412 a
BergerBN2	7.10 ab	2.84 b	0.958 ab	0.280 b
Germinaza	2.80 d	1.00 c	0.453 d	0.185 c
Germinaza 50%+Humus lombriz 50%	4.64 c	2.22 b	0.856 b	0.290 b
DMS	1.85	0.99	0.20	0.08

Where: Means with the same letter within columns are statistically equal, according to Tukey's test (p≤ 0.05), LSD= Least Significant Difference.

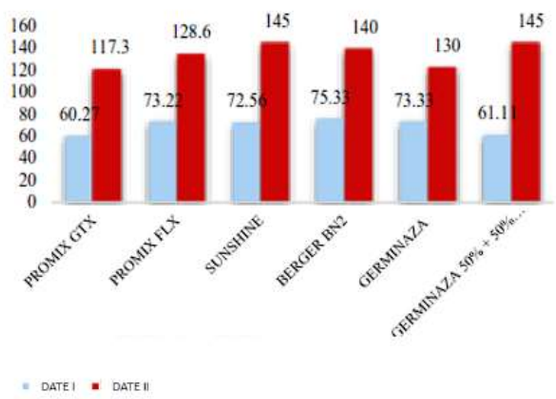
Table 2 Comparative test of means (Tukey at 0.05 probability) for the variables fresh weight of aerial part (PFPA), fresh weight of root (PFRA), dry weight of aerial part (PSPA), and dry weight of root (PSRA) in the six substrates evaluated



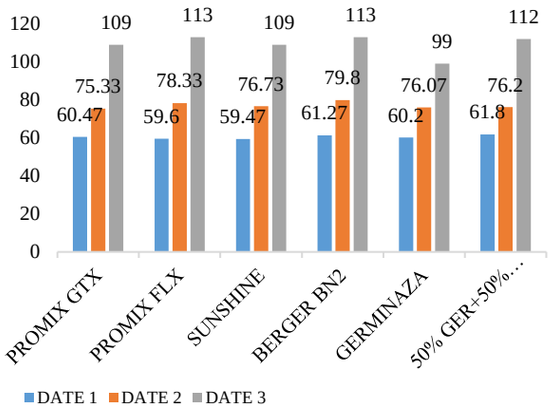
Graph 1. a) Variable seedling length (cm) and b) variable stem thickness (mm) in the six treatments evaluated



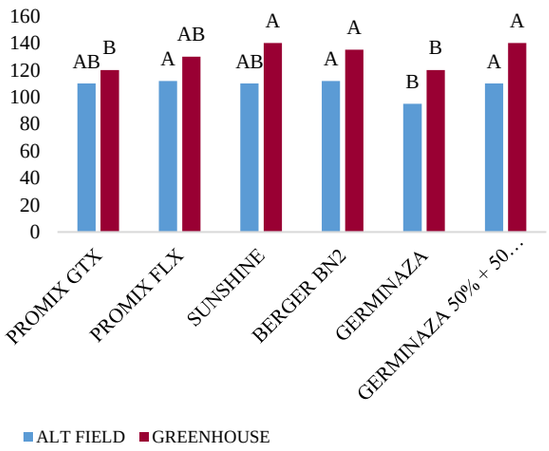
Graph 2 Variable number of leaves in the six treatments evaluated in Yahualica chile seedlings



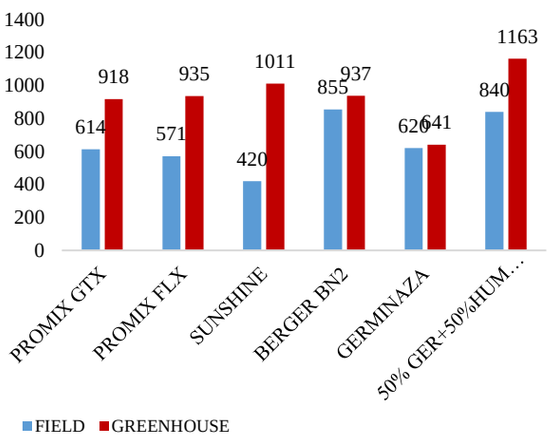
Graph 3 Variable plant height (cm) on two sampling dates for the six substrates evaluated under greenhouse conditions



Graph 4 Variable plant height (cm) during three dates in the six substrates evaluated in the field



Graph 5 Comparison of the variable plant height (cm) at harvest between greenhouse and field for the six treatments evaluated. Means with the same letter within columns are statistically equal



Graph 6 Variable fresh fruit weight (gr) at harvest for the six treatments evaluated under field and greenhouse conditions

References

Abad, M. y Noguera, P. (2000). Los sustratos en los cultivos sin suelo. En Manual del cultivo sin suelo. Universidad de Almería- Mundi-Prensa, Madrid, 137-183 p. URL: https://www.scirp.org/(S(351jmbntvnsjt1aadkozje))/reference/referencespapers.aspx?referenceid=1837630

Acosta-Durán, C. M.; Gallardo, C. S.; Kämpf, A. N. y Bezerra, F. C. (2008). Materiales regionales utilizados en Latinoamérica para la preparación de sustratos. Investigación Agropecuaria, 5(2): 93-106. URL: https://www.academia.edu/3244556/ Materiales regionales utilizados_en_Latinoamerica_para_la_preparación_de _ sustratos.

Andrade-Rodríguez, M; Ayala-Hernández, J.J; Alia-Tejacal, I; Rodríguez-Mendoza, H; Acosta-Durán, C.M. y López-Martínez, V. (2008). Efecto de promotores de la germinación y sustratos en el desarrollo de plántulas de papayo. *Revista de la Facultad de Agronomía. 25*(4): 617-635. URL: http://ve.scielo.org/scielo.php?script=sci_arttext&pid=S0378-78182008000400002 & lng=es&tlng=es.

Berrospe-Ochoa, E. A.; V. M. Ordaz-Chaparro; M. N. Rodríguez-Mendoza y R. Quintero-Lisaola. (2012). Cachaza como sustrato para la producción de plántula de tomate. Rev. Chapingo Ser. Hortic. 18: 141-156. URL: http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S1027-152X2012000100010&lng=es&tlng=es.

Bowman, D.C., Paul, J.L. (1983). Understanding of container media vital knowledge for growing successful plants. Pacific Coast Nurseryman and Garden Supply Dealer. March Issue. https://doi.org/:10.21930/agrosavia.manual.7405682

Díaz, P. J. C; Granberry, D.M.; Germishuizen, P. (2008). Transplant growth and stand establishment of bell pepper (Capsicum annuum L.) plants as affected by compost-amended substrate. Acta Hort. 782: 223- 228. https://doi.org/: 10.17660/ActaHortic.2008.782.26

Domínguez, J.; Lazcano, C. y Gómez-Brandón, M. (2010). Influencia del vermicomposta en el crecimiento de las plantas. Aportes para la elaboración de un concepto objetivo. Acta Zoológica Mexicana. Número Especial 2: 359-371. https://doi.org/:10.21829/azm.2010.262900

Fernández, B. C; Urdanet, N. y Silva, W. (2006). Germinación de semillas de tomate (*Lycopersicum esculentum* Mill.) Cv Río Grande sembradas en bandejas plásticas, utilizando distintos sustratos. Rev. Fac. Agron.23 (2): 188-196.URL: http://ve.scielo.org/scielo.php?script=sci_arttext&pid=S0378-78182006000200006&lng=es&tlng=es.

García, C.O.; Alcántar, G.G.; Cabrera, R.I.; F. Gavi R. y V. Volke, H. (2001). Evaluación de sustratos para la producción de *Epipremnum aureum* y *Spathiphyllum wallissi* cultivadas en maceta. Terra latinoamericana 19 (3): 249-258. URL: https://www.redalyc.org/articulo.oa?id=57319306

García, H. 2004. Modificación al sistema de clasificación climática de Koppen. Instituto de geografía, Universidad Nacional Autónoma de México. Quinta edición. Núm. 6. URL: http://www.publicaciones.igg.unam.mx/index.php/ig/catalog/book/83

Gomes, L. A. A.; Rodríguez, A. A.; Collier, L. S. e Feitosa, S. S. (2008). Produjao de mudas de alface em substrato alternativo com adubajao. Horticultura Brasileira. 26: p 359-363. URL: https://www.scielo.br/j/hb/a/TN4PQz85nZkyGWDB4psmvkL/?format=pdf&lang=pt

Grigatti, M.; Giorgioni, M. E. and Ciavatta, C. (2007). Compost-based growing media: influence on growth and nutrient use of bedding plants. Bioresour. Technol. 98:3526-3534. https://doi.org/: 10.1016/j.biortech.2006.11.016

Guimarães, V.F.; M.M. Echer y K. Minami, (2002). Métodos de produção de mudas, distribuição de matéria seca produtividade de plântulas de beterraba. Hortic. Bras. 20 (3), 505-509. https:// doi.org/:10.1590/S0102-05362002000300022

Guzman, M. and Sánchez, A. (2003). Influence of nitrate and calcium increments on development, growth and early yield in sweet pepper plants. Acta Hort. 609: 207-211. https://doi.org/:10.17660/ActaHortic.2003.609.29

Hartmann, H. y Kester, D. (2002). Plant propagation. Principles and practices. Prentice Hall. New Jersey. P 880. URL: https://books.google.com.mx/books/about/Hartmann_and_Kester_s_Plant_Propagation.html?id=SNtXAAAAYAAJ&redir_esc=y

Herrera, F.; Castillo, J. E.; Chica, A. F.; L. López Bellido (2008). Use of municipal solid waste compost (MSWC) as a growing medium in the nursery production of tomato plants. Bioresource Technol. 99 (2): 287–296. https://doi.org/:10.1016/j.biortech.2006.12.042. Epub 2007 Mar 6. PMID: 17344043.

Larqué-Saavedra, A.; Martín-Mex, R.; Nexticapan-Garcéz, A; Vergara-Yoisura, S. y Gutiérrez-Rendón, M. (2010). Efecto del ácido salicílico en el crecimiento de plántulas de tomate. Rev. Chapingo Ser. Hortic. 16 (3): 183-187. URL: http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S1027-152X2010000300006&lng=es&tlng=es.

Leskovar, D.I. (2001). Producción y ecofisiología del trasplante hortícola. Buena Vista, Saltillo, Coahuila. Curso. P 1-24. URL: https://es.scribd.com/document/25499246/Produccion-y-ecofisiologia-del-trasplante-horticola

López-Baltazar, J.; Méndez-Matías, A.; Pliego-Marín, L.; Aragón-Robles, E. y Robles-Martínez, M. L. (2013). Evaluación agronómica de sustratos en plántulas de chile 'onza' (*Capsicum annuum*) en invernadero. *Revista mexicana de ciencias agrícolas*, 4(6):1139-1150. https://doi.org/:10.29312/remexca.v0i6.1278

López-Moctezuma, H.; Ferrera-Cerrato, R.; Farias-Larios, J.; Aguilar-Espinosa, S.; F. Bello, Ma. del R; López-Aguirre, J.G.. (2005). Micorriza arbuscular, bacillus y sustrato enriquecido con vermicomposta en el desarrollo de plantas de papayo. Terra Latinoamericana 23 (4): 523-531. URL: https://www.redalyc.org/pdf/573/57311146011.pdf

Maguire, J.D. (1962). Speed of germination-aid in selection and evaluation for Seedling Emergence and Vigor. Crop Science 2 (2): 176-177. URL: https://acsess.onlinelibrary.wiley.com/doi/10.2135/cropsci1962.0011183X000200020033x

Momirovic, N.; Misovic, M.; Cvetkovic, R. (2000). Effect of different substrates on sweet pepper (*Capsicum annuum* L. cv. Macvanka) transplants quality in organic farming production. Acta Hort. 533: 135-139. https://doi.org/:10.17660/ActaHortic.2000.533.15

Moreno-Reséndez, A.; Gómez-Fuentes, L.; Cano-Ruíz, P., Martínez-Cueto, V.; Reyes Carrillo, J. L.; Puente-Manríquez, J. L. y Rodríguez-Dimas, N. (2008). Genotipos de tomate en mezclas de vermicompostas: arena en invernadero. Terra Latinoamericana 26 (2): 103-109. URL: http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S0187-57792008000200002&lng=es&tlng=es.

Muñoz, R. J. J. (2002). Acondicionamiento nutritivo de plántulas de tomate y pimiento en semillero y su respuesta postransplante. Tesis doctoral Universidad de Almería. España. URL: https://dialnet.unirioja.es/servlet/tesis?codigo=222718

Nesmith, D.S.; Duval, J.R. (1998). The effect of container size. HortTechnology. 8(4): 495-498. URL: https://swfrec.ifas.ufl.edu/docs/pdf/veg-hort/transplant/trans_cs1.pdf

Noguera, P.; Abad, M.; Puchades, R.; Maquieira, A.; Noguera, V. (2003). Influence of particle size on physical and chemical properties of coconut coir dust as container medium. Communications in Soil Science and Plant Analysis 34(3-4): 593–605. https://doi.org/:10.1081/CSS/120017842

Ocampo, M. J.; Caballero, M. R.; y Tornero, C. M. A. (2005). Los sustratos en cultivos hortícolas y ornamentales. En: Agricultura, Ganadería, Ambiente y Desarrollo Sustentable. Tornero C. M. A., Silva G. S. E., Pérez A. R. Y Bonilla F. N. (Eds.). Publicación especial de la Benemérita Universidad Autónoma de Puebla, México. pp. 55-73. ISBN: 968 863 913 3.

Oliverio T. S. (2014). Evaluación de cinco sustratos para la producción en vivero de palo blanco (*Tabebuia donnell-smithii Rose*). Tesis de Grado. Universidad Rafael Landívar, Facultad de Ciencias Ambientales y Agrícolas. Licenciatura en Ingeniería Forestal con énfasis en Silvicultura y Manejo de Bosques. Guatemala. P 68. URL:http://biblio3.url.edu.gt/Tesario/2014/06/22/Tut-Maynor.pdf

Ortega-Martínez, L. D.; Sánchez-Olarte, J.; Díaz-Ruiz, R. y Ocampo-Mendoza, J. (2010). Efecto de diferentes sustratos en el crecimiento de plántulas de tomate (*Lycopersicum esculentum* Mill). Ra Ximhai, 6(3):365-372. https://doi.org/:10.35197/rx.06.03.2010.05.1o

Parra-Terraza, S., Salas-Núñez, E.; Villareal-Romero, R.; Hernández-Verdugo, S. y Sánchez-Peña, P. (2010). Relaciones nitrato/ amonio/ urea y concentraciones de potasio en la producción de plántulas de tomate. Rev. Chapingo Ser. Hortic. 16 (1): p 37-47. URL: http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S1027-152X2010000100006lng=es&tlng=es.

Pérez de la C., F.J.; Córdova, T. L.; Santacruz, V. A.; Castillo, G. F.; Cárdenas, S. E. y Delgado A. A. (2007). Relación entre vigor inicial, rendimiento y sus componentes en poblaciones de maíz chalqueño. Agric. Téc. Méx.33 (1):5-16. URL: https://www.redalyc.org/pdf/608/60833101.pdf

Pérez-Murcia, M. D.; Moral, R.; Moreno-Caselles, J.; Pérez-Espinosa, A.; Paredes, C. (2006). Use of composted sewage sludge in growth media for broccoli. Bioresour. Technol. 97 (1): 123-130. https://doi.org/:10.1016/j.biortech.2005.02.005

Ramos, G. F.; Aguilar-Rubalcaba, J. A.; López-Gutiérrez, M.A.; Ochoa-Fuentes M.; y Vázquez-Martínez, O. (2011). Efecto de abonos orgánicos en el rendimiento del cultivo de chile ancho (*Capsicum annuum L.*) sobre las características química del suelo de la parcela experimental. Investigación y Ciencia de la Universidad Autónoma de Aguascalientes 51: 3-9. URL: https://www.redalyc.org/pdf/674/67418397001.pdf

Reveles H., M. (2005). Apuntes del curso producción de plántula en invernadero. Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias, Centro de Investigación Regional Norte Centro, Campo Experimental Zacatecas. México. Documento de trabajo, p 6.

Rojas, J; Paniagua, F. (2015). Comportamiento agronómico de *Capsicum annuum* L., *Lycopersicon esculentum* M. y *Cucumis melo* L. bajo cultivo protegido hidropónico utilizando la solución universal de Steiner. Tesis Lic. San Carlos, Costa Rica, Tecnológico de Costa Rica. P 98. URL: https://repositoriotec.tec.ac.cr/bitstream/handle/2238/6413/comportamiento_agronomico_capsicum_annuum.pdf?sequence=1&isAllowed=y

Rosca, V (2009). Optimization of nitrogen concentration in the fertilization solution for production of seedlings in cell trays. Acta Hort. 807_91: 613-618. https://doi.org/: 10.17660/ActaHortic.2009.807.91

Velasco-Velasco,V. J.;Ferrera-Cerrato,R.; Almaraz-Suarez, J.J.(2001). Vermicomposta, micorriza arbuscular y *Azospirillum brasilense* en tomate de cáscara. Terra latinoamericana19 (3): 241-248. URL: https://www.redalyc.org/pdf/573/57319305.pdf

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Use of Agroindustrial Waste of Orange to Obtain: Bioalcohol, Essential Oils and Activated Carbon

Aprovechamiento de los Residuos Agroindustriales de la Naranja para la Obtención de: Bioalcohol, Aceites Esenciales y Carbón Activado

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Abstract

The generation of agro-industrial waste in recent years has caused a worldwide problem, which is why the concern arises to take advantage of its components to be used, sincesaid waste can be treated until the negative impact that its disposal could generate is reduced; turning them into a useful and value-added product. The objective of thisproject is to take advantage of the agro-industrial residuesof the orange to obtain bioalcohol, essential oils and activated carbon, beginning with the collection of theresidues in orchards and industries and through a pre-treatment to them, of which a part passes to a fermentation process with Saccharomyces cerevisiae and the 2nd. Part, it goes through an extraction process by hydrodistillation, the waste obtained from the previous processes is used to obtain activated carbon, in this way we generate a secondary product that benefits said process by significantly reducing any residue.

Agroindustrial Waste, Essential Oils, Activated Carbon, Bioalcohol.

Resumen

La generación de residuos agroindustriales en los últimos años ha provocado una problemática a nivel mundial, porlo que surge la inquietud de aprovechar sus componentes para ser utilizados, ya que dichos residuos pueden ser tratados hasta reducir el impacto negativo que sudisposición pudiera generar; convirtiéndolos en un producto útil y de valor agregado. El objetivo de este proyecto es aprovechar los residuos agroindustriales de la naranja para la obtención de bioalcohol, aceites esenciales y carbón activado, iniciando con la recolección de los residuos en huertas e industrias y mediante un pretratamiento a los mismos, de los cuales una parte pasa a un proceso de fermentación con Saccharomyces cerevisiae y la 2a. parte, pasa a un proceso de extracción por hidrodestilación, los desechos obtenidos de los procesos anteriores son usados para la obtención de carbón activado, de esta manera generamos un producto secundario que beneficia dicho proceso al disminuir significativamente cualquier residuo.

Residuo Agroindustrial, Aceites Esenciales, Carbón Activado, Bioalcohol.

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Introduction

Agro-industrial waste is generated by the different food and agricultural industries and is generally not of interest in the process that generates it. In recent years, several environmental problems have been linked to its generation, there is a benefit in implementing processes that allow an efficient and integral use of waste, so the concern arises to take advantage of its components to be used, since agro-industrial waste can be treated to reduce the negative impact that its disposal could generate; turning it into a useful and value-added product. In Nuevo León, orange production is 4.9% with 326,000 tonnes, and the waste generated by orange production is peel and syrup. Oranges contain fermentable sugars such as glucose, fructose and sucrose as well as insoluble polysaccharides.

Use the solid residues from the processes involved:

1. Those generated from the fermentation of oranges.
2. Those generated from the extraction of the essential oil.

Using them to obtain activated carbon and thus eliminate the waste in order to reduce the polluting effects caused by the accumulation of this waste.

In the disposal of citrus waste, between 45 and 60% of its weight is obtained as waste and is distributed in: peels (50 to 55% of the waste), peel (30 to 35%) and seeds (about 10%) (Garzón and González, 2012). This results in high quality residues during processing.

In 2014, the University of Antioquia demonstrated that orange peels are suitable for ethanol production because they contain high concentrations of soluble sugars; they are rich in soluble sugars and have a high content in sugars that can be used in the production of ethanol soluble sugars; they are rich in fermentable sugars such as glucose, fructose and sucrose.

Orange peels are particularly suitable for conversion to biofuels such as ethanol because of their polymer content and soluble and insoluble carbohydrate content, low lignin content and high concentrations of fermentable sugars such as glucose, fructose and sucrose.

Orange contains 9.35g/100g corresponding to sucrose, dextrose and levulose, protein 0.9g/100g, fat 0.1g/100g. The highly aromatic peel (exocarp) contains an essential oil rich in flavonoids.

Lignocellulosic orange residues produce activated charcoal by pyrolysis through impregnation by chemical dehydrating agents, carbonised at low cost, but with textural properties suitable for environmental uses.
Agro-industrial waste is usually produced in large quantities, so if it is not disposed of or managed properly, it can generate environmental pollution.

Our geographical area belongs to the citrus-growing region, being the orange the most consumed in the municipality of Cadereyta Jiménez, Nuevo León, mainly used in the food industry to obtain products such as juices or non-carbonated beverages.

Companies do not usually take full control of their waste, causing a serious problem for society such as the increase of greenhouse gases, soil contamination and the production of carbon footprint. Small companies on the other hand do not have the knowledge or resources to sustain a correct management and handling of waste, therefore, they discard orange peels, without having taken full advantage of the properties that they have, generating bad odour, generating bad smell, and even causing the waste to be discarded.

This generates a bad smell, an infestation of flies (or other animals) or even the incineration of the material, increasing global warming.

Hence, the need to carry out a sustainable project that allows an: Integral use of agro-industrial orange waste and use the total solid waste derived from the processes of oil extraction and alcoholic fermentation, for an application as activated carbon, as it has countless applications in the fields of: medicine, the biopharmaceutical industry and the environment.

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Development of the sections:

1. Collection of agro-industrial waste

The orange waste is collected from orchards, small juiceries in the municipality of Cadereyta.

2. Treatment of agro-industrial waste

The collected oranges are washed with potable water to remove unwanted matter. They are then sorted to discard the rotting material. They undergo a size reduction process to obtain squares of approximately 1 cm on each side, then they are dried in an oven at a temperature of 60°C for 72 hours until they are free of moisture and then ground to obtain a flour.

3. Alcoholic fermentation

A scheme is designed for the fermentation stage of the process, which allows varying the amount of yeast (0.10, 0.12, 0.15, 0.20) %, as well as the fermentation time (4, 5, 6 and 7) days respectively (see Table 1). In a container, 0.5 kg of previously crushed orange is added with 4 L of water, 0.1 to 0.2% of previously diluted yeast is added, an airlock valve is placed in the upper part of the container, the resting time for the fermentation process is 4 to 7 days, with a temperature range of 25°C. (See Table 1). After fermentation, the solids are separated (for later use) by a rapid filtration process and the filtered liquor is distilled by a simple distillation process to separate the alcohol.

4. Extraction of essential oils

A scheme for the extraction of essential oils by hydrodistillation is designed, with 60 g of sample and a time of 6 hr at 98°C, then filtered, the solid residues are dried at 60°C for 8 hr and then fermented. (See Table 2).

5. Obtaining activated carbon

For this stage a scheme is designed starting with the collection of the dry solid residues from the alcoholic fermentation and essential oil extraction processes, then we move on to chemical activation which is impregnation with an activating agent which in this case is phosphoric acid (H3PO4) at 30%, followed by agitation at 100°C for 2 hours followed by washing with distilled water and finally drying at 200°C for 4 hours (see Table 3). (See Table 3).

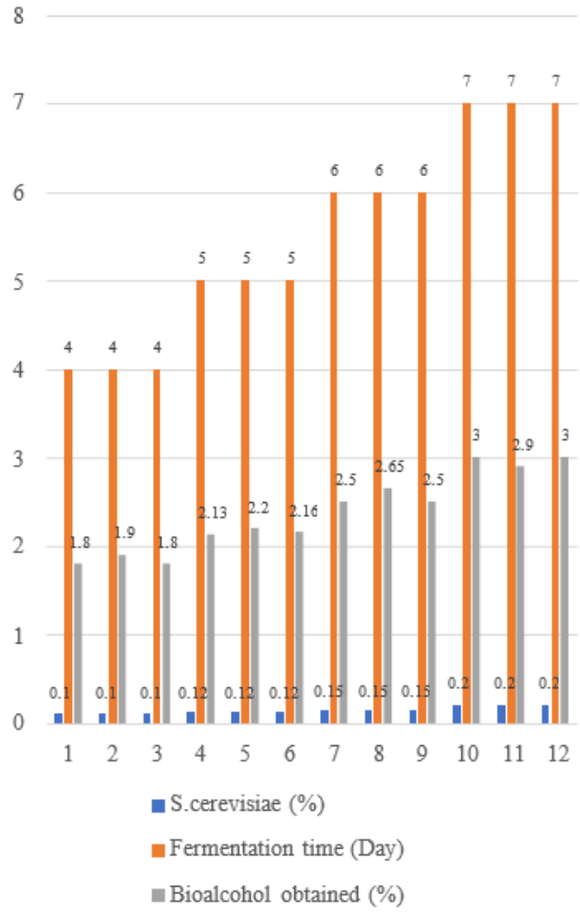

Figure 1 Obtaining bioalcohol

Ensayo (No.)	Residuo (Kg)	Agua (L)	Azucares (°Brix)	S.cerevisiae (%)	Tiempo de fermentacion (día)	Alcohol (%)
1	0.5	4	13	0.1	4.0	1.8
2	0.5	4	13	0.1	4.0	1.9
3	0.5	4	13	0.1	4.0	1.8
4	0.5	4	13	0.12	5.0	2.13
5	0.5	4	13	0.12	5.0	2.2
6	0.5	4	13	0.12	5.0	2.16
7	0.5	4	13	0.15	6.0	2.5
8	0.5	4	13	0.15	6.0	2.65
9	0.5	4	13	0.15	6.0	2.5
10	0.5	4	13	0.2	7.0	3.0
11	0.5	4	13	0.2	7.0	2.9
12	0.5	4	13	0.2	7.0	3.0

Table 1 Bioalcohol used in the production of variables

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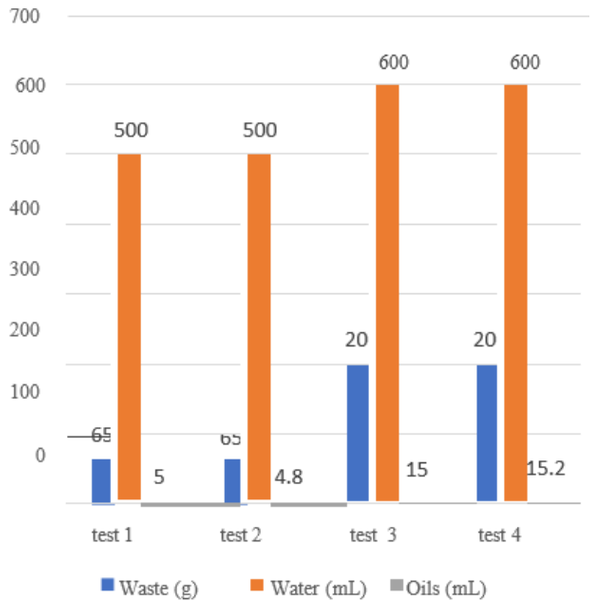

Figure 2 Oil production.

Essay	Orange waste (g)	Amount of water (mL)	Essential oils (mL)
1	65.0	500.0	5.0
2	65.0	500.0	4.8
3	200.0	600.0	15.0
4	200.0	600.0	15.2

Table 2 Variables used in the production of oils

Essay	Process waste (Kg)	Phosphoric acid 30% (L)	Distilled water (L)	Activated Charcoal (g)
1	0.1	0.250	0.4	45
2	0.1	0.250	0.4	48
3	0.1	0.250	0.4	47

Table 3 Variables used in the production of activated carbon

Methodology to be developed

1. Collection of agro-industrial waste

This starts with a plan to visit the small orchards and juice plantations in the municipality of Cadereyta, to establish the criteria for accepting their agro-industrial waste and to observe its viability for the processes, thus managing a permit to apply for it, for transfer to the processing site.

2. Treatment of agro-industrial wastes

This starts with the criterion of using oranges that are not in a state of putrefaction free of mud, leaves, animals, etc. They are then washed with potable water to remove unwanted matter.

They are then reduced in size to obtain squares of approximately 1 cm on each side, after which they are dried in an oven at a temperature of 60°C for 72 hours until they are free of moisture, and then ground to obtain a flour using an industrial blender.

3. Alcoholic fermentation

A scheme is designed for the fermentation process stage, which allows to vary the amount of Saccharomyces cerevisiae yeast (0.10, 0.12, 0.15, 0.20) %, as well as the fermentation time (4, 5, 6 and 7 days respectively) fermentation time (4, 5, 6 and 7) days, respectively (See Table 1)

In a biological reactor, 0.5Kg of previously crushed orange is added with 4L of water, 0.1 to 0.2% of previously diluted yeast is added, an airlock valve is placed at the top of the reactor, the resting time for the fermentation process is 4 to 7 days, in a heating bath with a temperature of 25°C. (See Table 1).

After fermentation, the solids are separated (for further use) by a rapid filtration process and the filtered liquor is distilled by a simple distillation process to separate the alcohol. The solid residues obtained from this process are dried in a Shel lab oven at 60°C for 8 hours.

4. Extraction of essential oils

A scheme is designed for the extraction of essential oils by hydrodistillation, in a 1000mL Florence flask, 400mL of water is added with 60 g of sample, which is previously cut and dried, in a time of 6 hrs. at 98°C, the content of essential oils is determined by the following process.is obtained by density difference with the help of a separating funnel.

The solid residue from this process is filtered and dried in a Shel lab oven at 60°C for 8 hours to be taken to the next process.

5. Obtaining activated carbon

For this stage a scheme is designed, starting with the collection of the dry solid waste from the alcoholic fermentation and essential oil extraction processes, it is weighed and we move on to chemical activation, which is by impregnation with an activating agent, in this case phosphoric acid (H3PO4) at 30%,

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Then it is taken to the carbonisation stage in the Termoscientific muffle at 380°C for 2 hours, followed by washing with distilled water in order to eliminate the remains of the acid until a neutral pH is reached, and finally it is dried at 60°C for 24 hours in the Shel lab oven.

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Conclusions

Agro-industrial wastes are organic in nature and are practically sorted at source, which facilitates their recycling, thus transforming "a problem into an opportunity".

The general objective is fulfilled with satisfactory results, however, one of the challenges of this research is to make the processes more efficient and to characterise the products from agro-industrial waste, in order to find their possible applications.

Therefore, we can conclude that through biotechnology it is possible to bioconvert agro-industrial orange waste by means of direct extraction processes (hydrodistillation), microbial transformation (fermentation) and/or chemical transformation (activated carbon) into commercial products with higher added value and greater impact, with the intention of improving environmental quality through technologies oriented towards a sustainable transformation of natural resources.

Therefore, it is concluded that, from the agro-industrial waste of the Cadereyta, Nuevo León orange, and according to the results observed:

With an initial pH between 3.0 and 4.0, adding yeast from 0.10 to 0.20% and extending the fermentation time allows us to obtain a greater amount of alcohol, which increases from 1.8 to 3.0% respectively; however, the search continues for an alternative methodology that will give us a higher percentage of alcohol, for its characterisation and future applications that will mainly benefit our community. In addition, the percentage of essential oils obtained is in a very favourable range (0.1 to 0.2%) per 100g of orange. To culminate the processes of this project, apart from obtaining alcohol and oils, it opens up the possibility of using the waste from the previous processes to obtain an activated carbon.

References

ARELLANO, C. (2015). Obtención de bioetanol a partir de materiales lignocelulósicos sometidos a hidrólisis enzimática. Universidad Veracruzana. Https://cdigital.uv.mx/bitstream/handle/123456789/46812/arellanoperalescarla.pdf?Sequence=2&isallowed= y (enero 2023)

BELTRAN, M. (2018). Estudio del contenido de azúcares totales en cáscaras de naranja para su uso en la producción de bioetanol. Universidad pública de navarra. https://academica-e.unavarra.es/xmlui/bitstream/handle/2454/38648/BELTRAN%20SI%C3%91ANI%2C%20MAGALY%20INES.pdf?sequence=1&isAllowed=y (enero 2023)

ESPINOZA B, Zingale D, Rubal- Peace G. Prevalence of medically unsupervised activated charcoal use a cause for concern in celiac disease?. J Am Pharm Assoc 2022;62:546-550. View abstract. Prevalence of medically unsupervised activated charcoal use a cause for concern in celiac disease? - PubMed (nih.gov) (febrero 2023)

GARCÍA, R.; GRANILLO, Y. (2017). "Evaluación de las condiciones operacionales en el proceso de preparación de carbón activo de cáscara de naranja valencia (Citrus Sinensis Linn Osbeck). Nicaragua: Universidad Nacional Autónoma de Nicaragua. Evaluación de las condiciones operacionales en el proceso de preparación de carbón activo de cáscara de naranja valencia (citrus sinensis linn obsbeck), laboratorios de química UNAN-Managua, II semestre 2016 - Repositorio Institucional UNAN-Managua(febrero 2023)

GARZÓN, J.; GONZÁLEZ, L. (2012). "Adsorción de Cr (VI) utilizando carbón activado a partir de cáscara de naranja". Colombia: Universidad de Cartagena. Adsorción de Cr (VI) a partir de cáscara de naranja y carbón activado obtenido de la misma (19.12.12).pdf (unicartagena.edu.co) (febrero 2023)

GUTIÉRREZ, W. (2020). Optimización del proceso de producción de etanol a partir de la cáscarade naranja aplicando metodología six sigma. Universidad nacional abierta y a distancia especialización en proceso de alimentos y biomateriales. https://repository.unad.edu.co/bitstream/handle/10596/37174/Optimizaciondelprocesodeproduccion deetanolapartirdelacascaradenaranjaaplicando metodologiasixsigma.pdf?sequence=1&isAllowed=y (enero 2023)

LLUMIQUINGA, J. (2018). Diseño deuna planta piloto para la producción de gel antibacterial. Universidad central del ecuador, facultad de ingeniería química. http://www.dspace.uce.edu.ec/bitstream/25000/17290/1/T-UCE-0017- IQU-027.pdf (enero 2023)

MEJÍA, L. F., & Martínez Correa, H. A., & Betancourt Gutiérrez, J. E., & Castrillón Castaño, C. E. (2007). Aprovechamiento del residuo agroindustrial del mango común (Mangifera indica L.) En la obtención de azúcares fermentables. Ingeniería y Ciencia, 3(6),41-62. Https://www.redalyc.org/articulo.oa?Id=83530603 (enero 2023)

PINZÓN-BEDOYA, M, & CARDONA A. (2008). "Caracterización de la cáscara de naranja para su uso como material adsorbente.". Bistua: Revista de la Facultad de Ciencias Básicas, 6(1), 1- 23. [fecha de Consulta 2 de Abril de 2022]. https://www.redalyc.org/articulo.oa?id=90312176003 (enero 2023)

QUIROZ, A. (2009). Utilización de residuos de cáscara de naranja para la preparación de un desengrasante doméstico e industrial. universidad internacional sek, Facultad de Ciencias Ambientales. https://repositorio.uisek.edu.ec/bitstream/123456789/407/1/Utilizaci%C3%B3n%20de%20residuos%20de%20c%C3%A1scara%20de%20naranja%20para%20la%20preparaci%C3%B3n%20de%20un%20desengrasante%20dom%C3%A9stico%20e%20in dustrial (enero 2023)

TEJEDA, L., Tejeda, C., Villabona, A., Alvear, M., Castillo, C., Henao, L., Marimon, W., Madariaga, N., & Taron, A. (2010, diciembre). Producción de bioetanol a partir de la fermentación alcohólica de jarabes glucosados derivados de cáscaras de naranja y piña. Asociación Colombiana de Facultades de Ingeniería. Https://www.acofi.edu.co/revista/Revista10/2009_I_51.pdf (enero 2023)

TREJO, M., Bravo, A., Venegas, J., & Islas, J. (2016, junio). Hidrólisis ácida de Citrus sinensis para la obtención de azúcares fermentables. Instituto Tecnológico Superior de Huichapan. Https://www.ecorfan.org/bolivia/researchjournals/Tecnologia_e_innovacion/vol3num7/Revista_de_Tecnologia_e_Innovaci%C3%b3n_V3_N7_6. pdf (enero 2023)

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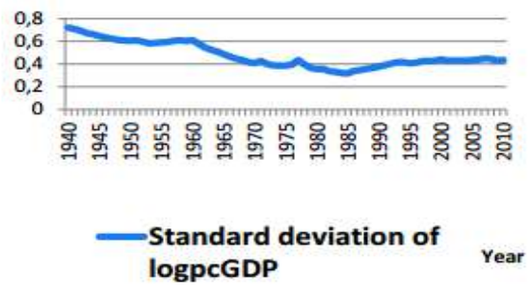

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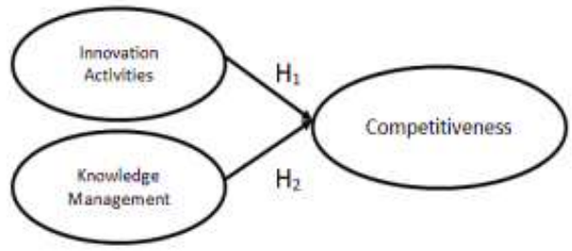

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