Colegio de Postgraduados

Economic evaluation of a business model of a *Vanilla planifolia* biofactory using BIT® bioreactors

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ABSTRACT

Objective: To evaluate the economic and financial viability for the implementation of a biofactory responsible for the *in vitro* propagation of *Vanilla planifolia* with TIB® bioreactors.

Design/methodology/approach: A completely randomized design was used. The data were processed with the IBM SPSS Statistics software (version 21). A Mann-Whitney U test was performed ($p \leq 0.05$). An Economic-Financial Evaluation was carried out, determining the main economic indicators: Profitability, Minimum Acceptable Rate of Return (MARR), Internal Rate of Return (IRR), Net Present Value (NPV) and Benefit-Cost Ratio (B/C R).

Results: TIB® temporary immersion bioreactors were used in this study. An average multiplication rate of 18.37 shoots per explant was obtained. When performing the economic-financial analysis of this agribusiness model over a five-year horizon, it yielded an Internal Rate of Return of 12.36%, a Benefit-Cost Ratio of 1.81, and a Net Present Value of MX$286,506.73 pesos (US$14,474.93), with a payback period of two years and seven months.

Limitations on study/implications: Using semi-solid culture media in the multiplication stage in vanilla decreases the production capacity and significantly lowers the profitability of the biofactory.

Findings/conclusions: The profitability of a biofactory for the production of vanilla depends on the multiplication rate, achieved in this case satisfactorily through the use of TIB®. This vanilla biofactory agribusiness model using bioreactors can be adopted by investors as an economic development strategy.

Keywords: biofactory, bioreactors, economic evaluation.

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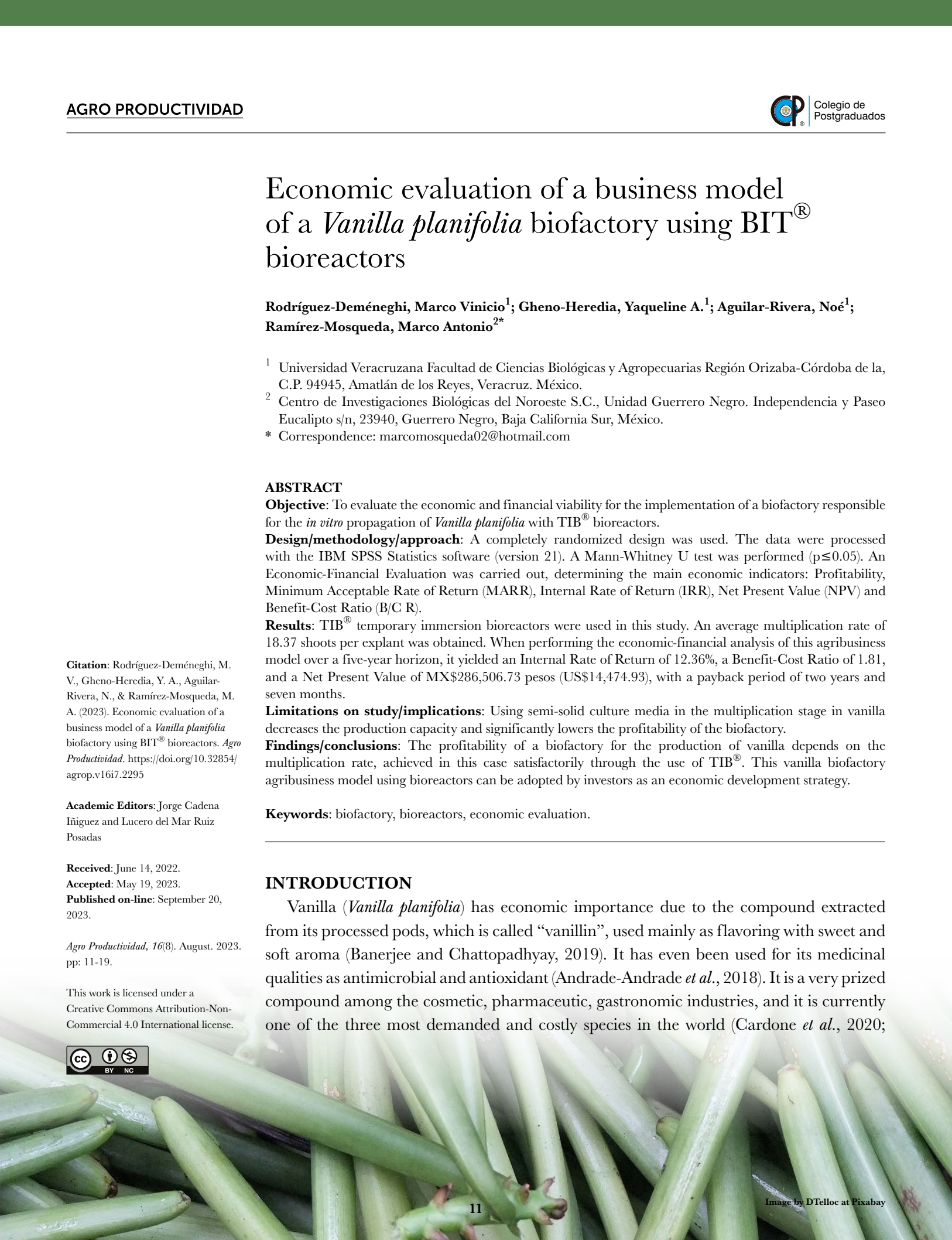
INTRODUCTION

Vanilla (*Vanilla planifolia*) has economic importance due to the compound extracted from its processed pods, which is called "vanillin", used mainly as flavoring with sweet and soft aroma (Banerjee and Chattopadhyay, 2019). It has even been used for its medicinal qualities as antimicrobial and antioxidant (Andrade-Andrade *et al.*, 2018). It is a very prized compound among the cosmetic, pharmaceutic, gastronomic industries, and it is currently one of the three most demanded and costly species in the world (Cardone *et al.*, 2020;



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Khoyratty et al., 2018); as a producing country, Mexico occupies the fourth place globally (FAOTAST, 2020). However, this species presents serious reproduction problems, because of the low germination percentage of its seeds (≤1 %) and the limited number of cuttings that can be obtained from an adult plant (Ramírez-Mosqueda and Iglesias-Andreu, 2016; Ramírez-Mosqueda and Bello-Bello, 2021).

Three methods are used in Mexico to propagate vanilla. 1) Cuttings or stem segments of 70-80 cm, 2) *In vitro* culture, and 3) Seeds. The first is the one used commercially for its propagation. In the vanilla-producing regions of Mexico, the cuttings are obtained from commercial plantations, since there are no nurseries devoted exclusively to their production (Santillán-Fernández, 2019). Convential propagation is slow, it requires much work and consumes a lot of time for the growth and development of the mother plant (Geetha and Shetty, 2000; Giridhar and Ravishankar, 2004). The second case refers to *in vitro* culture, which has been developed recently in greater proportion; these micropropagation techniques can attain a large amount of propagules in small spaces and in less time (Loyola-Vargas and Ochoa-Alejo, 2018). The third case, seeds, is limited by the low viability and germination rate of the seeds (Sasikumar, 2010).

In this sense, the micropropagation obtained from plant tissue culture (PTC) techniques has become the basis of a large industry of commercial plant propagation that involves hundreds of laboratories around the world. It is crucial to exploit the *in vitro* culture potential to multiply and supply the necessary amount of planting materials at a large scale (Abebe *et al.*, 2009). This technology can also be used to satisfy the market's demand for vanilla and to surpass the difficulties in alternative propagation methods.

Achieving the micropropagation of *V. planifolia* in a most efficient way has been attained through biotechnology, with the use of Temporary Immersion Bioreactors (TIB) (Ramírez-Mosqueda and Iglesias-Andreu, 2016). However, these scientific advances have not been used in the implementation of a biofactory, to supply producers from the Totonacapan region with commercial propagules. Biofactories can be defined as centers for the large-scale production of improved plants and seeds to obtain specimens with novel scientific techniques that guarantee their quality (CONACYT, 2016). Biofactories operate as productive businesses, which is why their development is based on the economic results from their sales (Suarez-Castellá *et al.*, 2009). In this sense, the implementation of a biofactory to cultivate vanilla using the TIB bioreactor would be an extremely efficient alternative in the commercial propagation of this valuable crop.

MATERIALS AND METHODS

Plant material

To conduct this research study, the methodology reported by Ramírez-Mosqueda and Iglesias-Andreu (2016), on the evaluation of different temporary immersion systems in the shoot proliferation phase, was replicated. *In vitro* plants of *V. planifolia* were used, obtained in the Plant Micropropagation Laboratory (Laboratorio de Micropropagación Vegetal, LAMICROVE) of the Facultad de Ciencias Biológicas y Agropecuarias Región Orizaba-Córdoba at the Universidad Veracruzana (UV) in Amatlán de los Reyes, Veracruz, Mexico.

Evaluation of two temporary immersion systems in the shoot proliferation phase

Nodal segments (1-2 cm of length) were cultivated in two temporary immersion systems: RITA® 100 ml (150×130 mm) and TIB® 940 ml (180×100 mm). In all the experiments, the MS (Murashige and Skoog, 1962) culture medium was used, added with 50 mg L^{-1} of cysteine hydrochloride, 100 mg L^{-1} of ascorbic acid, 30 g L^{-1} of sucrose. The pH of the medium was adjusted to 5.8±0.2 and the culture jars were sterilized in an autoclave at 1.5 kg cm^{-2} of pressure and 121 °C for 15 min. Then, 2.1 mg L^{-1} of benzyladenine (BA) was added and an immersion frequency of 2 min every 8 hours was used, with a volume of medium of 25 ml per explant. The cultures were incubated at a temperature of 25±2 °C, under radiation of 50 μmol m^{-2} s^{-1}, provided by LED lamps. For each immersion system, 50 explants were used (10 explants per jar with five repetitions). After six weeks of culture, the following variables were evaluated: number and length of shoots formed and number of leaves.

Rooting and acclimation

Individual shoots with 2 cm length were rooted in TIB bioreactors, using MS medium at 50% of its concentration, without plant growth regulators. The pH of the medium was adjusted to 5.8±0.2 and the culture jars were sterilized in an autoclave at 1.5 kg cm^{-2} of pressure and 121 °C for 15 min. The time, frequency of immersion, and incubation conditions were the same as those described before. For the acclimation, shoots with a height of 8-10 cm previously rooted *in vitro* were rinsed with running water and sown in (1:1 v/v) 1:1 peat moss mixture (Premier, Rivière-du-Loup, Canada) and agrolite® (Agrolita, Tlalnepantla de Baz, Mexico) as substrate, using trays of 50×30×5 cm. The seedlings were kept under greenhouse conditions (shade at 50%, relative moisture between 80-95%, temperature 28-32 °C), Nitrofoska® (N: 25 P: 10 K: 17) (PS, COMPO, Zapopan, Mexico) was applied as leaf fertilizer once per week, as well as irrigation with running water three times per week. When the plants reached 30 cm of height, they were transferred to individual containers with the same substrate.

Statistical analysis

A completely randomized design was used in these experiments. The data that were obtained from this experiment were processed with the IBM SPSS Statistics software (version 21). A Mann-Whitney U test was conducted ($p \leq 0.05$).

Economic-Financial Evaluation

An Economic-Financial Evaluation was carried out based on an agribusiness model described by Pavón (2012), where the implementation of a biofactory model is considered, establishing the main economic indicators: profitability, minimum acceptable rate of return (MARR), internal rate of return (IRR), net present value (NPV), and benefit-cost rate (B/C R) to understand the viability of this project.

RESULTS AND DISCUSSION

Evaluation of two different temporary immersion systems in the shoot proliferation phase

At six weeks of culture, significant differences were observed between the two temporary immersion systems evaluated (Table 1). The largest number of shoots/explants (18.37) was obtained in the BIT® bioreactor, followed by RITA® (12.62). However, longer shoots were obtained (1.67 cm) in the RITA® bioreactor compared to BIT® with 1.43 cm. No significant differences were observed for the number of leaves in BIT®.

The values represent the mean ±SE (standard error). Means with different letter are significantly different (U de Mann-Whitney ($p \leq 0.05$). Figure 1 shows the development of shoots/explants of *V. planifolia* in the temporary immersion bioreactor TIB®.

Based on the results obtained in our study, it was corroborated that the methodology proposed by Ramírez-Mosqueda and Iglesias-Andreu (2016) is efficient for the micropropagation of *V. planifolia*, using TIB® bioreactors. The TIB® bioreactor has been efficient for micropropagation of sugarcane (*Saccharum* sp.) (Lorenzo *et al.*, 1998), production of potato microtubers (*Solanum tuberosum* L) (Tapia *et al.*, 2020), and papaya (*Carica papaya* L.) (Gómez-Carrera, 2018). Ramos-Castellá *et al.*, determined that the RITA® bioreactor was efficient for the commercial micropropagation of *V. planifolia*. However, our study shows how TIB® generated higher biological yields, expressed in number of shoots. Ramírez-Mosqueda and Bello-Bello (2021) determined that shoots of

Table 1. Effect of two types of temporary immersion systems in the proliferation phase of *Vanilla planifolia* shoots at six weeks of culture, using the methodology proposed by Ramírez-Mosqueda and Iglesias-Andreu (2016).

Bioreactor Type	Number of shoots/explant	Shoot length (cm)	Number of leaves/ shoot
RITA®	12.62 ± 0.26^{b}	1.67 ± 0.09^{a}	1.85 ± 0.19^{a}
BIT®	18.37 ± 0.19^{a}	1.43 ± 0.10^{b}	1.77 ± 0.20^{a}

Values represent mean ± SE (standard error). Means with different letters are significantly different (Mann-Whitney U test ($p \leq 0.05$).

Figure 1. Result from the temporary immersion system in the shoot proliferation phase of *Vanilla planifolia* in TIB®: A) development of *V. planifolia* shoots, B) and C) specimens of *V. planifolia* established in bioreactor TIB® in acclimation stage.

V. planifolia generated in the SETIS™ bioreactor are taller in comparison with TIB®. However, in the multiplication phase, what is sought is to obtain the greatest number of commercial shoots. On the other hand, the TIB® bioreactor has a lower cost per unit than all bioreactors offered commercially. In this sense, Alamilla-Magaña *et al.* (2019) mention that the decrease in the initial cost of a biofactory is essential to ensure its success. Therefore, the TIB® bioreactors fulfill these qualities of being economic and functional.

Rooting and acclimation

On hundred percent rooting was achieved from the *V. planifolia* shoots using TIB® bioreactors. In addition, 100% of survival was attained during the acclimation process (Figure 1).

In vitro rooting using reduced culture means, without the addition of RCV, is an option that reduces the production costs (Alamilla-Magaña *et al.*, 2019). In this sense, our study was successful in this phase of micropropagation, which could be implemented in a biofactory. The acclimation phase is one of the most difficult in PTC, and therefore, its success reflects the efficiency of micropropagation protocols. The plants generated in TIS also present physiological advantages to those propagated in conventional systems (semi-solid), among them the stomatic functionality due to gas exchange (Georgiev *et al.*, 2014). In the present study, the efficiency of the protocol described is corroborated, with the attainment of 100% survival of the plants generated.

Economic-Financial Evaluation

To determine whether this agribusiness model of vanilla biofactory using TIB® bioreactors is feasible, an economic evaluation was carried out where the financial indicators of a private investment project were determined and where a source of financing from an external institution was contemplated. To start the operations of the biofactory, an initial investment of MX$ 605,380.07 is required, equivalent to US$ 30,585.10 (at an exchange rate of $19.7933 MX pesos per dollar, 26/05/22, BANXICO). In this investment, the construction of facilities was not considered, since a segment is contemplated for their rental within the operation expenses, although this investment includes equipment, materials, reagants, and assemblage of a greenhouse area for acclimation, required for the establishment of the biofactory. The fixed investment estimated is MX$ 440,380.07 (US$ 22,248.94), a deferred investment of MX$ 15,000.00 MX pesos (US$ 757.83), which is attributed to the legal construction of the company, as well as the specialized training on the operation of the TIS and working capital of MX$ 150,000.00 (US$ 7,578.32). It is expected that, for every productive year (in a horizon projected of 5 years), there will be an annual production of 20,000 vanilla seedlings and sales of MX$ 500,000.00 (US$ 25,261.07). The recovery period was achieved at two years and seven months after the project began. Table 2 shows the calculation memory used to define the financial indicators presented in this same study. A MARR of 10% was obtained, which is a minimal measure of profitability that was demanded from the

Table 2. Calculation memory to obtain financial indicators in a vanilla biofactory business model.

Concept	Unit of measurement	Calculation memory		Total (MX$)
		Quantity	Amount ($)	
Equipment	Batch	1	363,671.60	363,671.60
Reagents for propagation	Batch	1	47,590.16	47,590.16
Consumables	Batch	1	8,314.24	8,314.24
Area equipment	Batch	1	20,804.07	20,804.07
			TOTAL	$ 440,380.07
Other expenses				**Cost (MX$)**
Production and labor				209,531.56
Administration and sale				59,600.00
Financial expenses				18,000.00
Performance and sale price				**Total**
Multiplication rate		(units)		20,000
Unit price				$25.00

project and was obtained from the inflation rate for the year 2021, issued by Banco de México plus a risk premium (7.36+2.64=10). An IRR of 12.36% was obtained and this is equivalent to the interest rate provided to investors, this benefit percentage should be higher than the MARR and it reflects that the initial investment has been recovered. Regarding the NPV obtained of MX$286,506.73 (US$ 14,474.93), this criterion suggests that the project should be accepted since its net present value (NPV) is equal or higher than zero, where the NPV is the difference between all its revenues and expenditures expressed in present currency. After having conducted the corresponding operation for the calculation of the Benefit/Cost Rate (B/C R), the value obtained is positive and reaches $1.81, which indicates that for every peso invested, $0.81 cents of benefit will be obtained, where the percentage represents a profitability of 81%. The main indicators which express the economic yield of an investment were determined, and decisions can be made based on these to determine whether a project is viable or, if aplicable, to make a profitability assessment of such an investment. Chino (2011) points out that the most frequently used economic indicators are those that consider the value of money over time, among which there is: Minimum Acceptable Rate of Return (MARR); Internal Rate of Return (IRR); Net Present Value (NPV); and Benefit/Cost Rate (B/C R). In this sense, Baca (2001) mentions that the MARR is the real growth rate of the company above inflation, which is why in this project it is attained with the calculation. Meanwhile, Pavón (2012) indicates that the project gives a higher profitability than the minimum required profitability. The NPV considers the value of money over time; it is the numerical difference between the updated value of the benefits (utility) and the updated value of the costs at a specific updated rate, and when the numerical difference is positive, the investment is recommendable (Chino, 2011). Chino (2011) indicates that the B/C R value should be higher than zero, which indicates that the activity is generating utilities, so in this project it is fulfilled with this parameter.

CONCLUSIONS

Based on the results obtained in our study, the efficiency of the TIB® temporary immersion systems in the micropropagation of *V. planifolia* using the methodology by Ramírez-Mosqueda and Iglesias-Andreu (2016) was demonstrated. The use of TIB® bioreactors, which because of their acquisition cost that is rather lower than other types of temporal immersion systems, sets the path to be used in the assemblage of a commercial vanilla biofactory, which was the objective of this study. The results from the economic-financial evaluation indicators of a biofactory business model for *Vanilla planifolia* using TIB® bioreactors are positive, indicating the acceptability of the project based on an agribusiness model. The economic projections reflected the viability of this project, since it assures to be completely profitable based on the economic indicators. There are many research studies about the cultivation of vanilla, however, no studies have been conducted about the implementation of a vanilla biofactory company and the value of its costs in an investment project in this agricultural sector. This agribusiness model of a vanilla biofactory using TIB® bioreactors can be adopted by investors as an economic development strategy, considering the potential for growth in producing zones and in face of the need that producers have for quality plant material that is resistant to pests and diseases.

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