World Academy of Science, Engineering and Technology
International Journal of Mathematical and Computational Sciences
Vol:14, No:12, 2020

Isolation, Identification and Characterization of the Bacteria and Yeast from the Fermented Stevia Extract

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Abstract : Stevia (Stevia rebaudiana Bertoni) is a composite plant native to Paraguay. Stevia sweetener is derived from a hot water extract of Stevia (Stevia extract), which has some effects such as histamine decomposition, antioxidative effect, and blood sugar level-lowering function. The steviol glycosides in the Stevia extract are considered to contribute to these effects. In addition, these effects increase by the fermentation. However, it takes a long time for fermentation of Stevia extract and the fermentation liquid sometimes decays during the fermentation process because natural fermentation method is used. The aim of this study is to perform the fermentation of Stevia extract in a shorter period, and to produce the fermentation liquid in stable quality. From the natural fermentation liquid of Stevia extract, the four strains of useful (good taste) microorganisms were isolated using dilution plate count method and some properties were determined. The base sequences of 16S rDNA and 28S rDNA revealed three bacteria (two Lactobacillus sp. and Microbacterium sp.) and one yeast (Issatchenkia sp.). This result has corresponded that several kinds of lactic bacterium such as Lactobacillus pentosus and Lactobacillus buchneri were isolated from Stevia leaves. Liquid chromatography/mass spectrometory (LC/MS/MS) and High-Performance Liquid Chromatography (HPLC) were used to determine the contents of steviol glycosides and neutral sugars. When these strains were cultured in the sterile Stevia extract, the steviol and stevioside were increased in the fermented Stevia extract. So, it was suggested that the rebaudioside A and the mixture of steviol glycosides in the Stevia extract were decomposed into stevioside and steviol by microbial metabolism.

Keywords : fermentation, lactobacillus, Stevia, steviol glycosides, yeast
Conference Title : ICSRD 2020 : International Conference on Scientific Research and Development
Conference Location : Chicago, United States
Conference Dates : December 12-13, 2020