UTILIZATION OF SALTED WHEY FOR CULTIVATION OF SOME ECONOMICAL IMPORTANT MICROORGANISMS

By

Heba Mohammed Mahmoud Kanzy
B. Sc. Agric. Sci., (Food science), Fac. Agric., Cairo Univ., 2007

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APPROVAL SHEET

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ABSTRACT

Production of some Egyptian types of cheese gives huge amount of salted whey as a byproduct. Therefore, the present study was carried out to evaluate utilization of salted cheese whey as raw material for production of economically important products. There is an increased interest in careteniod as natural antioxidant pigments with many important biological activities which can reduce chronic diseases. A new yeast strain was isolated from salted cheese whey as carotenoid producing strain and identified according to its morphological, cultural and biochemical characteristics as *Rhodotorula* sp. For conclusive identification of *Rhodotorula* sp., the genus-specific 18s rRNA was carried out and identified as *R. mucilagenosa* (Y 17485). Its intracellular and extra cellular extracts showed no influence on mice survival. The identified strain and the reference strain *Rhodotoruola glutinis* (RCMB 028001) were grown to study the effect of NaCl concentration, incubation temperature, initial pH, incubation period, aeration and inoculum size on dry biomass and carotenoid production. The maximum biomass (13.95 g/l) and volumetric carotenoid production (6.544 mg/l) were scored by the reference strain *R. glutinis* after incubation for 120 hr at 30 °C and pH 6.6 in a medium containing 3% NaCl. The isolated strain showed its maximum biomass (9.02 g/l) in a medium containing 10% NaCl while the highest amount of volumetric carotenoid (5.044 mg/l) was obtained in a medium containing 6% NaCl after 120 hr incubatin at 30 °C and pH 6.6. The obtained result showed that the isolated strain *R. mucilagenosa* (Y 17485) will be a promising microorganism for commercial production of carotenoid.

Keywords: salted whey– *Rhodotoruola glutinis*- *R. mucilagenosa*- Carotenoids-factors affecting carotenoids production.

SUPERVISION SHEET

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DEDICATION

I dedicate this work to whom my heart felt thanks to my parents, my sister, my brothers and my friends for their patience, help and for all the support they lovely offered along the period of my post graduate studies.

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INTRODUCTION

Production of some Egyptian types of cheese gives huge amount of salted whey as a byproduct which contains approximately 4.5% lactose, 0.8% protein, 1% salt, and 0.1– 0.8% lactic acid. Annually, 110 million metric tons of whey are produced worldwide through cheese manufacture (Briczinski and Roberts, 2002). In the 1990s, whey production in Egypt increased steadily by about six percent annually and the amount produced in 2000 was 1,452,500 metric tons (Zhang *et al*., 2003). Salt whey is produced from Domiati, Ras and Cheddar cheese. The salt level in whey from Domiati cheese is about 8- 15%, whereas it is only about 2-5% in whey from Ras and Cheddar cheese making. Salt whey, unlike sweet whey, cannot be conveniently processed because of its high salinity level (Sanderson *et al*., 1996 and El-Tanboly *et al*., 2013). Moreover, it has a high biological (BOD) (35-40 g/l) and chemical oxygen demand (COD) (50 kg O_2/t permeate), which makes its disposal a problem (Zayed and Winter, 1995 and El-Tanboly *et al*., 2013). Most cheese manufacturing facilities in Egypt perform land spreading of salt whey. However, this practice increases the chloride levels of soil, and elevates the risk of crop damage (Awad *et al*., 2013). One possible application for salt whey is to use it as an ingredient in processed cheese (Code of Federal Regulations, 2003). Another application for salt whey is to use it as a whole medium for growth of milk clotting enzyme fungi (El-Tanboly *et al*., 2013). Many species of microorganism are known to produce a milk-clotting enzyme that can potentially substitute for calf rennet.

Rhizomucor pusillus, Rhizomucor miehei, Endothia parasitica and *Irpex lactis* are used extensively as sources of milk-clotting enzymes in the manufacture of cheese (O'Leary and Fox, 1974 and Kurutahalli *et al*., 2010).

Whey has been used in the production of single cell protein, alcoholic and non-alcoholic beverage, bakery products, and as an additive in dairy products. Gomashe *et al*., (2014) utilized whey protein and converting lactose to SCP using *Bacillus subtilis* strain. Ultrafiltration (UF) is generally used to separate proteins in whey from the permeate containing mostly lactose. Whey protein concentrate is an item of worldwide commerce due to its nutritious composition. Therefore, the use of whey in an inexpensive fermentation medium has long been of industrial interest. Lactose in whey is a suitable carbon source for many microorganisms. In addition, whey is rich in minerals and contains vitamins, which may provide valuable nutrients to stimulate cell growth and product formation. One of the promising ways to use lactose in whey is to use it as a low-cost carbon source for the production of carotenoid by fermentation (Aspasia and Anastassios, 2012)

Carotenoids are the most widespread natural lipid-soluble pigments with many important biological activities and industrial applications (Marova *et. al.*, 2004). Due to the growing demand for such compounds in pharmaceutical, cosmetic, food, and feed industries, there is increasing interest in carotenoids naturally obtained by biotechnological processes (Aksu and Eren, *2005*).

Carotenoids are important natural pigments, displaying yellow, orange, and red color, found widely in microorganisms and plants. Carotenoid pigments such as carotene and astaxanthin, are used as natural food colorants or feed additives in aquacultures. Several studies have shown that carotenoids combat various types of cancer and other diseases because of their antioxidant and/or provitamin A potential (Valduga *et al.,* 2009). Also, Marova *et al.*, (2012) reported that, carotenoid have important biological activity due to their provitamin A activity and properties resulting in potential health benefits such as strengthening the immune system and decreasing the risks of degenerative diseases. Furthermore, carotenoids have antioxidant action by neutralizing free radicals as electron donors. Thus, carotenoids as antioxidants prevent the damage caused on living cells by free radicals and ability to reduce chronic diseases.

Carotenoids are produced primarily by filamentous fungi and yeasts and by some species of bacteria, algae and lichens. Among microbial sources of carotenoids, specific yeasts strains take advantage of the utilization of the whole biomass, efficiently enriched for particular metabolites. Yeasts such as *Phafia rhodozyma* and *Rhodotorula glutinis* are of commercial interest (Martinez *et al*., 2009). Carotenoid production by fermentation can become industrially feasible if the cost of production can be minimized by use of cheap industrial by-products as nutrient sources. A number of studies have been carried out in recent years on the fermentation of various agricultural wastes (oats, wheat, barley, corn, rice, sugar cane