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METHODS FOR PREPARATION OF CASEIN HYDROLYSATES

Pratima Khandelwal*, Savitha N., Veeresh and Sruthi R[1].

Department of Biotechnology, New Horizon College of Engineering, Bangalore, India
[1] 2015 Passing out BE Biotechnology Student

ABSTRACT

Casein is the name for a family of related phosphoproteins (αS1, αS2, β, κ). These proteins are commonly found in mammalian milk, making up 80% of the proteins in cow's milk and between 20% and 45% of the proteins in human milk. Casein has a wide variety of uses, from being a major component of cheese, to use as a food additive, to a binder for safety matches. As a food source, casein supplies amino acids, carbohydrates, and the two inorganic elements calcium and phosphorus.

Key words: alcoholic fermentation, beverages**,** Saccharomyces

INTRODUCTION

Casein is the name for a family of related phosphoproteins (αS1, αS2, β, κ). These proteins are commonly found in mammalian milk, making up 80% of the proteins in cow's milk and between 20% and 45% of the proteins in human milk. Casein has a wide variety of uses, from being a major component of cheese, to use as a food additive, to a binder for safety matches. As a food source, casein supplies amino acids, carbohydrates, and the two inorganic elements calcium and phosphorus**.**

PREPARATION OF CASIEN

2 lit of skim milk was warmed (around 45°C), pH was adjusted to 4.6 using 0.1N HCL. The milk was thus coagulated and whey was supported out by filtration using a clean dried muslin cloth. The coagulum was collected, dried in hot air-oven at 60°C overnight. It was then powdered to a fine and uniform particle size in a pestle-mortar.

HYDROLISES OFCASIEN

500ml of Tris's buffer was prepared (PH8), 2. 5g casein was added to it along with 0.1g Trypsin. The dissolution was carried out using a magnetic stirrer at 35-40°C. When the PH becomes 5.2, temperature of the reaction mixture was increased to 80°C for a period of 10 minutes. The pH was then adjusted to 4.6 with dilute HCL after which the solution was centrifuged at 5000rpm for 10 minutes. The supernatant was collected, measured and accordingly $CaCl_2$ and ethanol were added at 1% and 50% (volume of a supernatant). The pH was finally adjusted to7.0 using NaoH. The solution was centrifuged at 5000rpm for 10 minutes. Pellet thus obtained were collected and dried at 50 °C.

EVALUATION OF GROWTH PARAMETERS

The product was serially diluted by taking 1ml of yoghourt containing Microorganism and suspending it into 9 ml of 0.85% saline, which was considered as 10-1. Further the dilution 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6}, 10^{-7}, 10^{-8}, 10^{-9}, 10^{-10}, 10^{-11} , 10^{-12}, 10^{-13}, 10^{-14}, 10^{-15} were prepared by suspending 1ml from each of the previous dilutions into 9ml of saline. Care was taken to have homogenous dilution by vortexing each test tube and then moving to next tube. 500µl of dilution was taken from 10^{-10}, 10^{-11}, 10^{-12}, 10^{-13}, 10^{-14}, 10^{-15} tubes and were pour plated using sterile MRS agar. The plates were allowed to solidify and incubated for 24-48 hours at 38.5°c to observe the colonies.

METHOD

The fermented milk product was developed using sterile of skim milk and different concentration of casein and caseino-phospho-peptide (CPPs). Fermentation was carried out by *Streptococcus Thermophilus* at 1% was incubated at 42°c at different intervals of time.
After the incubation the titre table products were analyzed for percentage of acidity by using the formula

$$\%\text{acidity} = \frac{\text{Normality of alkali x titer value (ml) x equivalent of acid x 100}}{\text{Volume of sample taken for estimation x 1000}}$$

Normality of alkali = 0.1
Equivalent of acid = 90
Volume of sample = 5 ml

Where:

Starter culture*: Streptococcus thermophilus,* Effect of CPPs and casiens on growth of *Streptococcus thermophilus.*
Incubation temperature: 42°C
CPP: casieno-phospho-peptides
Viable assay*: MRS agar (pour plating method) in Duplicate
Source of cultures: NDRI-Bangalore, Dairy Science College _Bangalore

METHOD

The fermented milk product was developed using sterile of skim milk and different concentration of casein and caseino- phosphor-peptide (CPPs).

Fermentation was carried out by *Bifido.bifidum* at 1% was incubated at 38.5°c at different intervals of time.

After the incubation the titer table products were analyzed for percentage of acidity by using the formula

$$\%\text{acidity} = \frac{\text{Normality of alkali x titer value (ml) x equivalent of acid x 100}}{\text{Volume of sample taken for estimation x 1000}}$$

Normality of alkali = 0.1
Equivalent of acid = 90
Volume of sample = 5 ml

Where:

Starter culture: *Bifido.bifidum*, Effect of CPPs and casiens on growth of *Bifido.bifidum*
Incubation temperature: 38.5°C

CPP: casieno-phospho-peptides

Viable assay*: MRS agar (pour plating method) in triplicate

Source of cultures: NDRI-Bangalore, Dairy Science College _Bangalore

METHOD

The fermented milk product was developed using sterile of skim milk and different concentration of casein and caseino- phosphor-peptide (CPPs).

Fermentation was carried out by Dahi at 1% was incubated at 39°c at different intervals of time.

After the incubation the titer table products were analyzed for percentage of acidity by using the formula

$$\%\text{acidity} = \frac{\text{Normality of alkali x titer value (ml) x equivalent of acid x 100}}{\text{Volume of sample taken for estimation x 1000}}$$

Normality of alkali = 0.1
Equivalent of acid = 90
Volume of sample = 5 ml

Where:

Starter culture: Dahi, Effect of CPPs and casiens on growth of Dahi
Incubation temperature: 39°C
CPP: casieno-phospho-peptides
Viable assay*: MRS agar (pour plating method) in Duplicate
Source of cultures: home made

METHOD

The fermented milk product was developed using sterile of skim milk and different concentration of casein and caseino -phospho- peptide (CPPs).

Fermentation was carried out by *lactobacillus bulgaricus* at 1% was incubated at 38.5°c at different intervals of time.

After the incubation the titer table products were analyzed for percentage of acidity by using the formula

$$\%\text{acidity} = \frac{\text{Normality of alkali x titer value (ml) x equivalent of acid x 100}}{\text{Volume of sample taken for estimation x 1000}}$$

Normality of alkali = 0.1
Equivalent of acid = 90
Volume of sample = 5 ml

Where:

Starter culture: *lactobacillus burglarious,* Effect of CPPs and casiens on growth of *lactobacillus bulgaricus.*
Incubation temperature: 38.5°C
CPP: casieno-phospho-peptides
Viable assay*: MRS agar (pour plating method) in Duplicate
Source of cultures: NDRI-Bangalore, Dairy Science College _Bangalore

METHOD

The fermented milk product was developed using sterile of skim milk and different concentration of casein and caseino- phosphor- peptide (CPPs).

Fermentation was carried out by *Lactobacillus .acidophilus* at 1% was incubated at 38.5°c at different intervals of time.

After the incubation the titer table products were analyzed for percentage of acidity by using the formula

$$\%\text{acidity} = \frac{\text{Normality of alkali x titer value (ml) x equivalent of acid}}{\text{Volume of sample taken for estimation x 1000}} 100$$

Where:

Starter culture: *lactobacillus acidophilus,* Effect of CPPs and casiens on growth of *lactobacillus acidophilus.*
Incubation temperature: 38.5°C
CPP: casieno-phospho-peptides
Viable assay*: MRS agar (pour plating method) in triplicate
Source of cultures: NDRI-Bangalore, Dairy Science College _Bangalore