Optimization of Invertase production using *Saccharomyces cerevisiae* MTCC 170 under varying cultural conditions

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ABSTRACT

A *Saccharomyces cerevisiae* MTCC 170 was obtained from MTCC. The effect of different production parameters such as pH, temperature, incubation time, carbon source, nitrogen source (organic and inorganic), inoculum concentrations, sucrose concentrations, metal ions, surfactants, amino acids, buffers, agricultural residues and agricultural residue concentration on invertase production by the isolated *Saccharomyces cerevisiae* MTCC 170 strain were studied. The enzyme production was assayed in submerged fermentation (SmF). Maximum invertase activity was found at pH 6, 30°C, 48 hours, sucrose, yeast extract (organic nitrogen), ammonium chloride (inorganic nitrogen), 2% inoculum concentration, 2% sucrose concentration, calcium chloride, poly ethylene glycol, methionine, citrate buffer and orange peel - 4%. A higher titre of invertase enzyme activity (0.43 ± 0.005 IU/ml) was obtained in the optimized production medium.

Keyword: invertase, optimization, *Saccharomyces cerevisiae* MTCC 170, SmF, SSF.

INTRODUCTION

Beta-fructofuranosidases (invertases) are enzymes that cleave "β-1,4-glucosidic linkage between "D-glucose and D-fructose molecules of sucrose by hydrolysis producing glucose and fructose. Beta-fructofuranosidases are intracellular as well as extracellular enzymes. It is used in the production of confectionery with liquid or soft centers, fermentation of cane molasses into ethanol, in calf feed preparation and also in manufacture of inverted sugars as food for honeybees [1]. Monosaccharide’s produced by microbial enzyme hydrolysis are more preferred because of high functionalities like similar taste as of sucrose and good bulking properties. Growth conditions have a great influence on invertase production capacity of *Saccharomyces cerevisiae*. The production of the extracellular invertase shows a cyclic behaviour that coincides with the budding cycle. The invertase activity increases during bud development and ceases at bud maturation and cell scission [3]. Invertase is classified in the GH32 family of glycoside hydrolases, that includes over 370 members and has been reported in plant, bacteria, yeast and filamentous fungi, as *Aspergillus ochraceus*, *Aspergillus niger*, *Aspergillus japonicas*, and *Thermomyces lanuginosus* (Alegre et al., 2009). Invertase is produced by *Saccharomyces cerevisiae* and *Saccharomyces carlsbergensis*, *Penicillium* and *Aspergillus* [5]. Cultivation conditions are essential in successful production of an enzyme and optimization of parameters such pH, temperature and media composition is important in developing the optimum fermentation conditions [6]. The extracellular thermostable amylases enzyme ranks first in terms of industrial point of view due to various industrial applications [5], particularly in starch processing industry. Industrially important enzymes have traditionally obtained from submerged fermentation (SMF) because of the ease of handling and greater control of environmental factors such as temperature and pH. The use of submerged culture is advantageous because of the easy of sterilization and process control is easier to engineer in these systems. Depending upon on the strain and culture conditions, the enzyme can be cultivable or inducible showing different production patterns [7]. The purpose of this work was to study the production of invertase by *Saccharomyces cerevisiae* MTCC 170 in optimized culture conditions for the production of invertase.
MATERIALS AND METHODS

Effect of pH on invertase production
The effect of optimum pH for invertase production by *Saccharomyces cerevisiae* MTCC 170 was determined by culturing the yeast in the production media with different pH. The experiment was carried out individually at various pH such as 2, 3, 4, 5, 6, 7, 8, 9 and 10. The enzyme assay was carried out after 48 hours of incubation at 30°C [8].

Effect of temperature on invertase production
Temperature plays an important role for the production of the invertase by *Saccharomyces cerevisiae* MTCC 170. The effect of temperature on invertase production was studied by incubating the culture media at various temperatures such as 10, 20, 30, 40, 50 and 60°C [9].

Effect of incubation time on invertase production
The effect of incubation time on invertase production by *Saccharomyces cerevisiae* MTCC 170 was determined by culturing the yeast in the production media. The experiment was carried out individually at various incubation times. They were 24, 48, 72 and 96 hours [17].

Effect of carbon sources on invertase production
To identify suitable carbon source for the invertase production by *Saccharomyces cerevisiae* MTCC 170, the following carbon sources were tested. The production medium containing sucrose, act as a carbon source. This sucrose was replaced by trehalose, maltose, galactose, mannose, fructose, glucose, raffinose, arabinose, lactose, xylose, starch, carboxyl methyl cellulose and sucrose. These carbon sources were tested individually at the concentration of 1% with dry substrate in the optimized production medium. The enzyme assay was carried out after 48 hours of incubation at 30°C [11].

Effect of organic nitrogen sources on invertase production
The invertase production by *Saccharomyces cerevisiae* MTCC 170 was optimized by supplementing different organic nitrogen sources. For this, six different organic nitrogen sources were tested individually at the concentration of 0.5% with dry substrate in the optimized carbon sources in production medium. The organic nitrogen sources used were yeast extract, glycin, peptone, gelatin, urea and casein. The organic nitrogen source that results maximum invertase production was then used for further study [12].

Effect of inorganic nitrogen sources on invertase production
The invertase production by *Saccharomyces cerevisiae* MTCC 170 was also optimized by supplementing different inorganic nitrogen sources. The different inorganic nitrogen sources used for the invertase production were ammonium nitrate, ammonium chloride, ammonium molybdate, potassium nitrate and sodium nitrate. These were tested individually at the concentration of 0.5% in production medium. The inorganic nitrogen source that results maximum invertase production was then used for further study [13].

Effect of different concentration of inoculum level on invertase production
Different concentration of inoculum level such as 0.5, 1.1, 1.5, 2, 2.5, 3 and 3.5% were tested for their ability to induce invertase production in the production medium [30].

Effect of various concentration of sucrose level on invertase production
Different concentration of sucrose level such as 0.5, 1, 1.5, 2, 2.5, 3 and 3.5% were tested for their ability to induce invertase production in the production medium [15].

Effect of metal ions on invertase production
In the present study to enhance invertase production ferrous sulphate, zinc sulphate, magnesium chloride, cobaltous chloride, manganese sulphate, sodium chloride and calcium chloride were tested as the source of metal ions. In this study they were incorporated individually into the production medium at the concentration of 0.02%. The effect was determined after 48 hours of incubation [16].

Effect of surfactants on invertase production
To identify the surfactants facilitating invertase production, five different surfactants were used for experimentation. They were Tween-20, Tween-80, SDS (Sodium Dodecyl Sulphate), Triton X-100 and PEG (Poly Ethylene Glycol). The selected surfactants were tested individually at the concentration of 0.2% in the optimized production medium [17].

Effect of amino acids on invertase production
In the present study to enhance invertase production different amino acids were used. The different amino acids sources used for the invertase production were asparagine, lysine, histidine, glutamicacid, arginine, proline, glutamine, leucine, alanine, methionine, serine, phenylalanine, aspartic acid, cysteine, tyrosine, tryptophan, isoleucine, and l-lysine. They were introduced into the production medium individually to determine the effect of amino acids on the invertase production [18].

Effect of buffers on invertase production
In the present study to enhance invertase production different buffers at pH-6 were tested. The different buffers used for the invertase production were sodium phosphate buffer, phosphate buffer, citrate buffer and potassium phosphate buffer. In this study optimized sources were incorporated into the production medium containing 0.1M buffer at pH 6. The effect was determined after 48 hours of incubation [19].

Effect of agricultural residue on invertase production
The invertase production by *Saccharomyces cerevisiae* MTCC 170 was optimized by supplementing different agricultural residues. For this, seven different agricultural residues were tested individually at the concentration of 2% with dry powdered substrate in the optimized production medium. The different agricultural residues used for the invertase production were pomegranate peel, sappota peel, pineapple peel, orange peel, lemon peel, grape peel and sugarcane bagasse [20].

Effect of different concentration of agricultural residue on invertase production
Different concentration of agricultural residue (orange peel) such as 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10% were tested for their ability to induce invertase production by *Saccharomyces cerevisiae* MTCC 170 in the production medium [21].

Invertase production media in Solid State Fermentation (SSF)
The (250 ml) Erlenmeyer flasks containing 40g of agricultural substrate (orange peel) and 20 ml of production medium containing (g/l): sucrose – 10.0g, yeast extract - 5.0g, ammonium chloride - 0.5g, calcium chloride - 0.2g, polyethylene glycol – 2.0g, methionine – 2.0g, 0.1M citrate buffer at pH-6 composition: citric acid - 9.5g and sodium citrate - 40.5g. The yeast *Saccharomyces cerevisiae* MTCC 170 was inoculated and incubated for 48 hours. At the end of the fermentation 50 ml distilled water was added to the fermented substrate and kept in shaker for 1 hour. Then it was centrifuged at 10,000 rpm for 5 minutes and the supernatant was used for further analytical work.
Invertase production media in Submerged Fermentation (SmF)
The (250 ml) Erlenmeyer flasks containing 40g of agricultural substrate (orange peel) and 50 ml of production medium containing (g/l); sucrose - 10g/ml, yeast extract - 5.0gm/L, ammonium chloride - 0.5gm/L, calcium chloride - 0.2gm/L, polyethyleneglycol - 2gm/L, methionine - 2gm/L, 0.1M citrate buffer at pH=6 composition: citric acid - 9.5gm/L and sodium citrate - 40.5gm/L. The yeast Saccharomyces cerevisiae MTCC 170 was inoculated individually and incubated for 48 hours.

RESULT
Effect of pH on invertase production
The effect of different pH on invertase production after 48 hours of incubation period at 30°C showed maximum amount of invertase production at pH 6.0 (0.21 ± 0.005). The minimum invertase production was recorded at pH 4.0 (0.13 ± 0). The invertase production was not recorded at pH 2, 3, 9 and 10 (Fig. 1).

Effect of temperature on invertase production
Among the various temperatures tested, the maximum invertase production was obtained at 30°C temperature (0.32 ± 0.005 IU/ml). On the other hand, the minimum amount of invertase production was observed at temperature 10°C (0.01 ± 0.005 IU/ml) (Fig. 2).

Effect of various incubation intervals on invertase production
The effect of different kinds of incubation time was tested on invertase production. The maximum amount of invertase production was observed in 48 hours incubation time (0.21 ± 0.005 IU/ml). The minimum amount of invertase production was obtained in 96 hours of incubation time (0.09 ± 0.002 IU IU/ml) (Fig. 3).

Effect of carbon sources on invertase production
The effect of carbon sources on invertase production by Saccharomyces cerevisiae MTCC 170 after 48 hours of incubation period at 30°C is given in (Fig. 4). Here the maximum invertase production was recorded in sucrose (0.21 ± 0.005 IU/ml) supplemented medium. The minimum invertase production was recorded in lactose (0.01 ± 0.005 IU/ml) added medium. The invertase production was not recorded at xylose and trehalose.

Effect of organic nitrogen sources on invertase production
The effect of different kinds of organic nitrogen sources on invertase production after 48 hours of incubation period at 30°C showed maximum amount of enzyme production in yeast extract (0.24 ± 0.005 IU/ml) supplemented medium and minimum amount of invertase production in urea (0.04 ± 0.005 IU/ml) supplemented medium (Fig. 5).

Effect of inorganic nitrogen sources on invertase production
The effect of different kinds of inorganic nitrogen sources on invertase production after 48 hours of incubation period at 30°C showed maximum amount of enzyme production in ammonium chloride (0.25 ± 0.005 IU/ml) supplemented medium and minimum amount of invertase production in potassium nitrate (0.11 ± 0.005 IU/ml) supplemented medium (Fig. 6).

Effect of different concentration of inoculum on invertase production
The initial inoculum level in the invertase media is a critical factor in fermentation process. The maximum invertase activity was registered at the 2% (0.31 ± 0.005 IU/ml) of inoculum level. On the other hand, the minimum amount of invertase production was observed at 3.5% of (0.20 ± 0.005 IU/ml) inoculum level (Fig. 7).

Effect of different concentration of sucrose on invertase production
The effect of different concentration of sucrose on invertase production after 48 hours of incubation period at 30°C showed maximum amount of enzyme production in 2% (0.31 ± 0.005 IU/ml) supplemented medium and minimum amount of invertase production in 3.5% (0.04 ± 0.005 IU/ml) supplemented medium (Fig. 8)

Effect of metal ions on invertase production
Among the tested metal ions, the maximum amount of enzyme production was recorded in calcium chloride (0.12 ± 0.004 IU/ml) added medium. Followed by this, magnesium sulphate (0.120 ± 0.005 IU/ml) was the second best metal ions on invertase production, whereas the minimum amount of invertase production was observed in manganese sulphate (0.06 ± 0.005 IU/ml) supplemented medium (Fig. 9).

Effect of surfactants on invertase production
The effect of different kinds of surfactants was tested on invertase production after 48 hours of incubation period at 30°C. Among the tested surfactants, the maximum amount of enzyme production was recorded in Poly ethylene glycol (0.16 ± 0.005 IU/ml) added medium. The minimum amount of invertase enzyme production was recorded in Tween-20 (0.06 ± 0.003 IU/ml) supplemented medium (Fig. 10).

Effect of amino acids on invertase production
The effect of different kinds of amino acids was tested on invertase production after 48 hours of incubation period at 30°C. Among the tested amino acids, the maximum amount of enzyme production was recorded in methionine (0.26 ± 0.005 IU/ml) added medium. The minimum amount of invertase enzyme production was recorded in I-lysine (0.03 ± 0.002 IU/ml) supplemented medium (Fig. 11).

Effect of buffer on invertase production
The effect of different kinds of buffers was tested on invertase production after 48 hours of incubation period at 30°C. Here the maximum invertase production was recorded in citrate buffer (0.18 ± 0.005 IU/ml) supplemented medium. The minimum invertase production was recorded in sodium phosphate buffer (0.12 ± 0.005 IU/ml) added medium (Fig. 12).

Effect of agricultural residue on invertase production
Different agricultural by products such as pomegranate peel, sappota peel, pineapple peel, orange peel, lemon peel, grape peel and sugarcane bagasse were tested for the production of invertase enzyme. The maximum invertase production was recorded in orange peel (0.30 ± 0.006) supplemented medium. The minimum invertase production was recorded in sugarcane bagasse (0.05 ± 0.011 IU/ml) added medium (Fig. 13)

Effect of different concentration of agricultural residue on invertase production
The effect of different concentration of agricultural residue (orange peel) on invertase production after 48 hours of incubation period at 30°C showed maximum amount of enzyme production in 4% (0.48 ± 0.011 IU/ml) supplemented medium and minimum amount of invertase production in 8% (0.15 ± 0.005 IU/ml) supplemented medium (Fig. 14).

Invertase production in solid state and submerged fermentation
In solid state fermentation, invertase production recorded by Saccharomyces cerevisiae MTCC 170 was 0.35 ± 0.01 IU/ml. In Submerged fermentation, invertase production recorded by and
Saccharomyces cerevisiae MTCC 170 was 0.43 ± 0.005 IU/ml (Fig. 15).

Figure 1. Effect of pH on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 5. Effect of organic nitrogen sources on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 2. Effect of temperature on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 6. Effect of inorganic nitrogen sources on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 3. Effect of incubation time on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 7. Effect of different concentration of inoculum level on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 4. Effect of carbon sources on invertase production by Saccharomyces cerevisiae MTCC 170

Figure 8. Effect of various concentration of sucrose level on invertase production by Saccharomyces cerevisiae MTCC 170
DISCUSSION
Production of invertase is largely dependent on initial pH of the fermentation medium. The effect of initial pH on enzyme production by *Saccharomyces cerevisiae* MTCC 170. Maximum invertase production of $0.21 \pm 0.005$ IU/ml was obtained at pH 6 and minimum invertase production of $0.13 \pm 0$ IU/ml was recorded in pH 4.0 by *Saccharomyces cerevisiae* MTCC 170. Similarly Persike *et al* also reported similar results, significant growth rate was observed at pH 5.5 however maximum product rate was noted at initial pH 6. It means that although growth is...
more favoured at pH 5.5, but as far as invertase production is concerned, pH 6 is best. Whereas, Kim et al reported transfructosylating enzyme from Bacillus macerans EG-6 the best enzyme yield at pH 7 and the minimum invertase production was reported at pH 4. Carbon source, pH value, temperature, presence of inducers, medium additives, aeration and growth time have been reported to be important parameters in optimizing enzyme production by Immanuel et al. (2006) and among these, pH was of major interest by Juhasz et al. (2004).

In the present study the effect of temperature on invertase production by Saccharomyces cerevisiae MTCC 170 was studied. Incubation temperature is one of the critical factors that have a profound effect on the production of invertase Saccharomyces cerevisiae MTCC 170 gave a maximum invertase production at 30°C. Maximum invertase production of 0.32 ± 0.005 IU/ml reported at 30°C and minimum invertase production of 0.01 ± 0.005 IU/ml was recorded at 10°C by Saccharomyces cerevisiae MTCC 170. Similar results have been shown by Shafiq et al. (2004) for invertase production using Saccharomyces cerevisiae KRI1 the maximum production of invertase was obtained when incubation temperature was maintained at 30°C.

In the present work the invertase production was studied by varying the incubation time from 24-96 hours for Saccharomyces cerevisiae MTCC 170. Maximum amount of invertase production of 0.21 ± 0.005 IU/ml was recorded in 48 hours and minimum invertase production of 0.09 ± 0.002 IU/ml was recorded in 96 hours incubation time by Saccharomyces cerevisiae MTCC 170. Ul-Haq and Ali, (2005) reported similar result by Saccharomyces cerevisiae for invertase production it reached a maximum after 48 hours of incubation. Poonawalla et al. (1965) investigated that invertase titers reached the maximum at 120 hour with both the organisms of Penicillium chrysogenum and other fungi.

The different carbon source such as trehalose, maltose, galactose, mannose, fructose, glucose, raffinose, arabinose, lactose, xylose, starch, carboxyl methyl cellulose and sucrose were reported for invertase production by Saccharomyces cerevisiae MTCC 170. Maximum amount of invertase production of 0.21 ± 0.005 IU/ml was recorded in sucrose and minimum invertase production of 0.01 ± 0.005 IU/ml was recorded in lactose by Saccharomyces cerevisiae MTCC 170. Kim et al. (2000) also investigated that sucrose was good carbon source for invertase production by Bacillus macerans-EG-6. Uma et al. (2010) also stated the similar result, sucrose as best carbon source for invertase production by Aspergillus flavus. Carbohydrates are an excellent source of carbon, oxygen, hydrogen and metabolic energy. They are frequently present in the media at a concentration of 0.5-30%.

After carbon source, the next major compound in the media is the nitrogen source. The presence of appropriate concentration of carbon and nitrogen source greatly influences the production metabolites/desirable product. Therefore, it is necessary to find out suitable carbon and nitrogen sources. The different substrates containing carbon source, nitrogen source have a major effect on the yeast to synthesize invertase (Suresh et al., 2012).

In the present study the nitrogen sources are tested, Maximum amount of invertase production of 0.24 ± 0.005 IU/ml was recorded in yeast extract (organic nitrogen source) and minimum invertase production of 0.04 ± 0.005 IU/ml was recorded in urea (organic nitrogen source) by Saccharomyces cerevisiae MTCC 170. Whereas, Shafiq et al. (2002) tested the nitrogen source peptone, it gave the maximum invertase production for Saccharomyces cerevisiae GCB-K5. The nitrogen constituent has an important influence on the invertase production because there is a strong correlation between nitrogen equilibrium and productivity of cells [4].

Maximum amount of invertase production of 0.25 ± 0.005 IU/ml was recorded in ammonium chloride (inorganic nitrogen source) and minimum invertase production of 0.11 ± 0.005 IU/ml was recorded in potassium nitrate (inorganic nitrogen source) by Saccharomyces cerevisiae MTCC 170. Qureshi et al. (2012) reported the similar result, that yeast extract was best nitrogen source for invertase production by Muco geophillus EFRL 03. Nitrogen sources and their concentrations have major biological effect on enzyme yield because sucrose metabolism shows a specific physiological response to the presence of nitrogen source reported by Nakano et al. (2000).

In the present investigation effect of various concentration of inoculum concentration on invertase production was studied in Saccharomyces cerevisiae MTCC 170 was studied. Maximum invertase production of 0.31 ± 0.005 IU/ml by was registered at 2% inoculum level and minimum amount of invertase production of 0.20 ± 0.005 IU/ml was registered in 3.5% inoculums level by Saccharomyces cerevisiae MTCC 170. Whereas, Shafiq et al. (2003b) stated that maximum invertase production was obtained with 1.0ml of inoculum 4% in Saccharomyces cerevisiae KR18. Whereas, Uma et al. (2010) reported that higher invertase production was obtained from 3% of inoculum level on the Aspergillus flavus. Above and below optimal level of inoculum size, a decrease in enzyme activity was achieved.

In the present study effect of different concentration of sucrose concentration on invertase production was studied in Saccharomyces cerevisiae MTCC 170. Maximum invertase production of 0.31 ± 0.005 IU/ml by was registered in 2% sucrose concentration and minimum amount of invertase production of 0.04 ± 0.005 IU/ml was registered in 3.5% sucrose concentration by Saccharomyces cerevisiae MTCC 170. Suresh et al. (2012) stated that maximum sucrose concentration was obtained at 30g/L of invertase production by Saccharomyces cerevisiae 3090. At low concentration of fructose and glucose, the products of sucrose hydrolysis by invertase, induced the expression of an invertase coding gene in Saccharomyces cerevisiae as reported by Ozcar et al. (1997). Higher concentrations of sucrose in fermentation medium induce catabolite repression of yeast invertase by Ul-Haq et al. (2003).

In the present work to enhance invertase production ferrous sulphate, zinc sulphate, magnesium chloride, cobaltous chloride, manganese sulphate, sodium chloride and calcium chloride were tested as the source of metal ions by Saccharomyces cerevisiae MTCC 170 was studied. Maximum amount of invertase production of 0.12 ± 0.004 IU/ml was recorded in calcium chloride and minimum invertase production of 0.06 ± 0.005 IU/ml was recorded in manganese sulphate by Saccharomyces cerevisiae MTCC 170. MK. Kim et al. (2000) also reported that magnesium chloride was the significant metal ions for invertase production by Bacillus macerans-EG-6.

In the present investigation, effect of surfactants on invertase production by Saccharomyces cerevisiae MTCC 170 was studied. Maximum amount of invertase production of 0.16 ± 0.005 IU/ml was recorded in poly ethylene glycol and minimum invertase production of 0.06 ± 0.003 IU/ml was recorded in Tween-20 by Saccharomyces cerevisiae MTCC 170.

In the present study, effect of amino acids on invertase production by Saccharomyces cerevisiae MTCC 170 was studied. Maximum
amount of invertase production of 0.26 ± 0.005 IU/ml was recorded in methionine and minimum invertase production of 0.03 ± 0.002 IU/ml was recorded in L-lysine by Saccharomyces cerevisiae MTCC 170. In the present work, various buffers were used to stabilize the pH on invertase production by Saccharomyces cerevisiae MTCC 170 was studied. Maximum amount of invertase production of 0.18 ± 0.005 IU/ml was recorded in citrate buffer and minimum invertase production of 0.12 ± 0.005 IU/ml was recorded in sodium phosphate buffer by Saccharomyces cerevisiae MTCC 170. In the present investigation, seven agricultural residues such as pomegranate peel, sappota peel, pineapple peel, orange peel, lemon peel, grape peel and sugarcane bagasse have been used as substrates. Maximum amount of invertase production of 0.30 ± 0.006 IU/ml was recorded in orange peel and minimum invertase production of 0.05 ± 0.011 IU/ml was recorded in sugarcane bagasse by Saccharomyces cerevisiae MTCC 170. Uma et al. (2010) stated that the pomegranate peel was found to be the best substrate for invertase from a Cladosporium cladosporioides in SmF. Several agro-industrial residues such as sugarcane bagasse, sugar beet pulp/husk, orange bagasse, oil cakes, apple pomace, grape juice, grape seed, coffee husk, wheat bran, cereals, straw, leaves, corncobs etc. have been used as substrates. In most parts of our country, these materials are used only as animal feeds while a large quantity is left on farmlands to be decomposed by microorganisms such as bacteria and fungi (Pandey et al., 2000). In the present study effect of different concentration of agricultural residues on invertase production was studied in Saccharomyces cerevisiae MTCC 170. Maximum invertase production of 0.43 ± 0.005 IU/ml by was registered in 4% sucrose concentration and minimum amount of invertase production of 0.11 ± 0.005 IU/ml was registered in 8% sucrose concentration by Saccharomyces cerevisiae MTCC 170. It was concluded that from economic point of view Saccharomyces cerevisiae MTCC 170 was optimized in various production parameters like pH, temperature, carbon source, nitrogen source (Organic and Inorganic), sucrose concentration, surfactants, metal ions, inoculum size, incubation time amino acid, buffer, and agricultural residue concentration. So it can be used for invertase production on cheaper and more easily available resources than on expensive and refined media. More over this study gives us a hint as well as the microbial wealth of invertase producing yeast which can be harnessed for biotechnological processes.

CONCLUSION

The yeast (Saccharomyces cerevisiae MTCC 170) capable of producing invertase was obtained from MTCC. The optimum cultural conditions for the production of invertase was found to be at pH 6, 30°C, 48 hours, sucrose, yeast extract (organic nitrogen), ammonium chloride (inorganic nitrogen), 2% inoculum concentration, 2% sucrose concentration, calcium chloride, poly ethylene glycol, methionine, citrate buffer and orange peel - 4%. A higher titre of invertase enzyme activity (0.43 ± 0.005 IU/ml) was obtained in the optimized production medium.

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