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# Valorization of Pineapple Peels through Single Cell Protein Production Using *Saccharomyces cerevisiae* NCDC 364

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#### **Abstract**

**Background and objective:** Pineapple peels contain significant quantities of carbohydrates, which can be used as cheap raw materials for production of commercially important products through fermentation. The aim of this study was to use this feed stock for the cultivation of *Saccharomyces cerevisiae* NCDC 364 and its use as single cell protein.

**Material and methods:** The single cell protein was produced using discarded pineapple peels and *Saccharomyces cerevisiae* NCDC 364. Optimization of bioprocess variables (temperature, pH, incubation period, carbon source and nitrogen source) affecting single cell protein production was carried out using classical "one factor at a time" approach. The harvested cells from optimized media were screened for amino acid content using high-performance thin-layer chromatography.

Results and conclusion: The Saccharomyces cerevisiae NCDC 364 produced maximum single cell protein in pineapple peel based media, compared to non-optimized media. The "one factor at a time" approach showed that the maximum biomass production was achieved at optimized levels of temperature of 25°C, pH of 5, incubation period of 120 h, carbon source of 1% sucrose and nitrogen source of 0.5% beef extract. The amino acid profiling of the harvested biomass using high-performance thin-layer chromatography analysis revealed that tryptophan included a comparatively higher concentration of 6.52%, followed by threonine (3.25%). Results of this study suggest that easily available raw materials such as fruit peels offer cost-effective substrates for production of commercially important microbial proteins for alarming global issues linked to protein malnutrition.

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#### 1. Introduction

Exponential growth of the world population has developed global problems associated with penury, starvation and protein deficiency. Production of proteins has been a subject of various research over the last few decades. The human need to explore alternative sources of dietary proteins with superior nutraceutical properties has resulted in scientific focuses on single cell proteins (SCP) of the microbial origins [1]. Use of SCP as food and feed additive is truly regarded as a green process since it extends a possibility of valorizing agro-wastes in protein production [2]. Up-to-date, a wide variety of agro-waste substrates have been used for cost-effective production of SCP using **SCP** and fungi. Of chief producing microorganisms, yeast continues to dominate the microbial list as they are majorly composed of high protein contents and low nucleic acid contents, making them ideal as protein supplements [3-7]. Yeast biomass is generally considered as safe for use as foods and feed additives because of its nontoxicity and easy digestibility. The presence of substantial quantities of vitamins, minerals, malic acids and lysines as well as high protein contents makes yeast biomasses ideal candidates for animal feed additives [8]. Yeast cells include a significant role in animal health as its use in feed enhances yeast immunomodulatory effects and thus is important in aquaculture and poultry productions [9]. Based on the literatures, SCP represents an attractive protein supplement with satiation properties that can improve appetite regulation in obese people [10].

The widening gap between the supply and demand for dietary proteins can be bridged by increasing the production of foods and supplementing the existing resources with novel foodstuffs. Development of novel protein sources such as fish protein concentrates, SCPs, soybean proteins and insect proteins has significantly contributed to decrease world protein deficiency [7]. In previous studies, animal based proteins such as fish protein concentrates have been investigated for cardiovascular diseases (due to the high saturated fats and cholesterol consumptions), bone disorders (bone reabsorptions due to sulfur-containing amino acid (AA) linked to animal proteins) and other physiological system diseases [11]. Vegetable proteins are incomplete proteins as they lack one or more essential AAs (EAAs) [12]. This highlights the significance of SCP as an ideal protein supplement for foods and feed uses [13]. The SCP is a dried microbial biomass protein used as protein supplement in human or animal foods. In addition to its high protein contents, SCP contains fats, carbohydrates, nucleic acids, vitamins and minerals. Presence of EAAs such as lysine and methionine in SCP, which are limited in most plants and animals, advances them to foods from plant and animal foods [14]. Microbial protein production is independent of seasonal variations and climatic fluctuations, making their production sustainable all the year. The nutritive profile of SCP comprises of low-fat contents, accelerated protein/carbohydrate ratios and high-protein contents with a wide spectrum of AAs [15]. Flexibility in production process and amenability of SCP production using waste substrates make SCP foods superior sources [14]. Production of SCP from low-cost agricultural and industrial wastes is a feasible and financially viable process which is very important as it provides a sustainable solution to the management of these wastes and the environmental pollution caused by their disposals. Although extensive studies have been carried out on production of SCP from yeasts, the idea of scaling up the production process still faces several difficulties [4,5]. Optimization of bioprocess variables and screening of alternative media components to replace commercial nutrient supplements can make the process economically feasible [16].

The major objective of the current study was to valorize high carbohydrate pineapple peels in SCP production using *Saccharomyces* (*S.*) *cerevisiae*. The present study used a fermentative approach for the production of SCP using pineapple peel wastes and optimization of bioprocess variables affecting fermentation. This study used an acid hydrolysis method for releasing fermentable sugars from pineapple peels and replacing complex media components with simple hydrolysates. The novelty of this study relies on the optimization of bioprocess variables for enhancing SCP production using acid hydrolyzed pineapple peel media, which was not widely discussed in previous studies. Use of high-performance thin-layer chromatography (HPTLC) method for deducing EAAs in SCP was less investigated in other studies.

#### 2. Materials and methods

#### 2.1. Media components and production strains

Discarded pineapple peels were collected from local fruit markets in Coimbatore, Tamil Nadu, India. Lactose, dextrose, maltose, starch, sucrose, peptone, sodium nitrate, ammonium acetate, ammonium sulfate, agar, yeast extract and beef extract were purchased from HiMedia Laboratories, Mumbai, India. All the media components were of analytical grades. Solvents were purchased from Merck, Germany. The *S. cerevisiae* NCDC 364 strain was purchased provided by the National Dairy Research Institute, Karnal (Haryana), India. The strain was subcultured in yeast extract peptone agar (pH 6.0), including 0.3% (w v<sup>-1</sup>) of yeast extract, 1.0% (w v<sup>-1</sup>) of peptone, 2.0% (w v<sup>-1</sup>) of dextrose, 2.0% (w v<sup>-1</sup>) of agar and 100 ml of distilled water (D.W.), for further use.

#### 2.2. Proximate nutritive composition of pineapple peels

Ash and moisture contents of pineapple peels were analyzed using A.P.H.A protocol [17]. Crude fiber and lipid contents of the wastes were analyzed using A.O.A.C method [18]. Protein contents of the pineapple wastes were assessed using Lowry method [19]. Total sugar and reducing sugar contents of the pineapple wastes were analyzed using anthrone [20] and DNS methods [21], respectively.

#### 2.3. Substrate preparation

Collected pineapple peels were washed several times with D.W. to remove adhered dirt. These were dried using hot-air oven at 50°C for 48 h and then ground into fine powders and stored in air-tight containers at room temperature until further use [22]. For substrate preparation, pineapple peel powders (8 g) were steam sterilized at 121°C for 20 min. Sterile D.W. was added to pretreated materials to make a final volume of 200 ml followed by boiling at 80°C for 30 min. The mixture was filtered using cheese cloth and acid hydrolysis was carried out using HCl (1% v v<sup>-1</sup>) at 121°C for 30 min. Moreover, pH of the hydrolysate was adjusted to 7 using 1 M of NaOH. Neutralized pineapple peel hydrolysate (PPH) was used instead of D.W. for the preparation of fermentation media [23].

# 2.4. Inoculum preparation and fermentation for SCP production

The SCP production in pineapple peels was carried out using method described by Phaff et al. [24]. One loopful of S. cerevisiae NCDC 364 was inoculated into 10 ml of yeast extract peptone broth media in 100-ml conical flasks and incubated in a shaker incubator at 150 rpm for 24 h at 25°C. This media was used as inoculum for SCP production. The fermentation study was carried out in 250 ml conical flasks, containing the media. Two different media were used to assess their feasibility for SCP production. Temperature control and agitation (150 rpm) were carried out using shaker incubator system (New Brunswick 126 R model, Eppendorf, India). Media 1 Basal medium included basal media components (10.0 g of D-glucose; 5.0 g of (NH<sub>2</sub>)<sub>2</sub>SO<sub>4</sub>; 1.0 g of KH<sub>2</sub>PO<sub>4</sub>; 0.5 g of MgSO<sub>4</sub>.7H<sub>2</sub>O; 0.1 g of NaCl and 0.1 g of CaCl<sub>2</sub>; pH 5.5) in 1000 ml of D.W. Media 2 included PPH based media (Peel Hydrolysate Media) and the basal media components without glucose (pH 5.5) dissolved in 1000 ml of PPH instead of D.W. The production media was sterilized at 121°C for 15 min. The sterilized media were inoculated with a 24 h old culture of S. cerevisiae NCDC 364 (1%) with a concentration of  $3.3 \times$ 10<sup>3</sup> CFU ml<sup>-1</sup> and incubated at 20°C for 5 days. Extraction

of yeast biomass from fermentation broth was carried out through harvesting cells by centrifugation at  $5000 \times g$  for 20 min at room temperature [25]. The harvested biomass was dried at  $80^{\circ}$ C overnight until reaching a constant weight. The protein content of biomass was calculated using Lowry et al. method [19].

#### 2.5. Optimization of the SCP production

Effects of various bioprocess variables on maximizing SCP production were assessed using classical "one factor at a time" approach. Briefly, 100 ml of the PPH were used instead of D.W. for each optimization. All experiments were carried out in triplicate and the level of various factors was adjusted to the range while other factors were adjusted to a constant level. Biomass (g 1-1) and protein (%) contents for SCP production were calculated for all samples of the experiments and expressed in mean ±SD (standard deviation) of the triplicates. The SCP production at various pH levels (4, 5, 6, 7 and 8), incubation temperatures (25, 35, 45, 55 and 65°C) and incubation times (24, 48, 72, 96, 120 and 144 h) was assessed. The pH was adjusted using 5 M of NaOH and 5 M of HCl. Effects of carbon source supplementation on SCP production were assessed by adding 1% (w v-1) various carbon sources of glucose, lactose, maltose, fructose and sucrose alone into the production media. Effects of various nitrogen sources on SCP production were assessed by adding 0.5% (w v<sup>-1</sup>) sodium nitrate, peptone, yeast extract, ammonium nitrate, beef extract and urea alone into the production media, as described previously. Combined effects of the optimized parameters were studied after sorting out the ideal variables and their respective levels with significant effects on overall SCP yields.

#### 2.6. Amino acid profiling

The AA profiling of the SCP was carried out using HPTLC. The harvested yeast biomass was dried at 80°C for 3 h, digested with 6 M of aqueous hydrochloric acid and dried under vacuum. The powdered sample was dissolved in D.W. Then, 1 µl of the acid digested sample solution was loaded onto a pre-coated silica gel (60F254 TLC) plate (5 × 10 cm; Merck, India) and analyzed using CAMAG-LINOMAT 5 instrument. A mixture of *n*-butanol-acetic acid-water (3:1:1) was used as mobile phase. Plates were sprayed using ninhydrin reagent and dried and documented photo-documentation chamber (CAMAG-REPROSTAR 3) under visible light and UV (254 nm). Furthermore, plates were scanned at 500 nm using CAMAG-TLC Scanner 3 using WIN CATS v.1.3.4 Software. Peak areas of the samples were compared to standard AAs (set in groups of four) and quantified [26].

#### 2.7. Statistical analysis

All experiments were carried out in triplicate. The standard error values were displayed as Y error bars in the graphs. The Analysis of variance statistical analysis was carried out using Minitab v.17.1.10 Statistical Software. Significant differences were reported with a 95% confidence level ( $P \le 0.05$ ).

#### 3. Results and discussion

#### 3.1. Proximate nutritive composition of pineapple peels

Proximate nutritive compositions of the pineapple peels are represented in Table 1. Abundant quantities of the total sugars and appreciable quantities of the reducing sugars make these sugars ideal candidates for SCP production. Quantity of the total sugars (39.23% ±0.17 of dry weight) reported in this study supports feasibility of the pineapple peels as substrates for SCP production. This result was similar to that in orange peels (39.66%) used for SCP production, as reported by previous studies [27]. In this study, results from the nutritive characterization were similar to those in other studies with minor variations [28,29]. In a previous study on hybrid varieties of pineapple, an increased total sugar content of 42.3% (dry weight of peels) was recorded, which was higher than that recorded in the current study  $(39.23\% \pm 0.17)$  [29]. In that study, a reducing sugar content of 28.8% ±0.13 (dry weight of peels) was observed in pineapple peels treated with cellulose 4 h prior to vinegar production. This was higher than that seen in the current study  $(25.86\% \pm 0.12)$  [30]. In a previous study on use of raw pineapple pulps for ethanol production, a reducing sugar content of 22.5% (w w<sup>-1</sup>) was observed [31]. These differences could be attributed to the differences in types of cultivars, geographic locations of cultivations and seasonal variations [29].

**Table 1.** Nutritive parameters (in percentage of dry weight) of pineapple peels

| Parameter       | Percentage of dry weight |
|-----------------|--------------------------|
| Total sugars    | $39.23 \pm 0.17$         |
| Reducing sugars | $25.86 \pm 0.12$         |
| Crude fiber     | $13.12 \pm 0.23$         |
| Protein         | $11.56 \pm 0.01$         |
| Fat             | $10.65 \pm 0.19$         |
| Ash             | $10.6 \pm 0.07$          |
| Moisture        | $84.13 \pm 0.07$         |

Data represents the mean of triplicates  $\pm$  standard deviation.

#### 3.2. Screening of media for SCP production

Production of yeast biomass protein from fruit wastes basically involves the substantial use of fermentable sugars in production media by yeast cells for AA biosynthesis followed by protein accumulation [32]. Therefore, the harvested dried biomass contains a substantial quantity of EAAs. In the present study, of the two different media screened for SCP production, the highest biomass and protein contents were achieved in Peel Hydrolysate Media (Table 2); possibly due to the availability of excess quantities of fermentable sugars from pineapple peels. These results were similar to results from other studies on SCP production from fruit based media and commercial media [14,33-35].

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**Table 2.** Single Cell Protein production in original and optimized medium

| Media                | Parameter                            | Concentration           | Biomass yield (g l <sup>-1</sup> ) | Percentage of protein in dried biomass (w w-1) |
|----------------------|--------------------------------------|-------------------------|------------------------------------|--|
| Basal medium         | Glucose                              | 10 g l <sup>-1</sup>    | $3.15 \pm 0.11$                    | $10.76 \pm 0.03$                               |
|                      | (NH2)2SO4                            | 5 g l <sup>-1</sup>     |                                    |  |
|                      | $KH_2PO_4$                           | 1 g l <sup>-1</sup>     |                                    |  |
|                      | MgSO <sub>4</sub> .7H <sub>2</sub> O | 0.5 g l <sup>-1</sup>   |                                    |  |
|                      | NaCl                                 | $0.1 \text{ g } 1^{-1}$ |                                    |  |
|                      | CaCl <sub>2</sub>                    | 0.1 g l <sup>-1</sup>   |                                    |  |
|                      | pН                                   | 5.5                     |                                    |  |
|                      | Incubation period                    | 120 h                   |                                    |  |
| PHM                  | Glucose                              | 10 g l <sup>-1</sup>    | $4.04 \pm 0.03$                    | $14.76 \pm 0.23$                               |
| (before              | (NH2)2SO4                            | 5 g l <sup>-1</sup>     |                                    |  |
| optimization)        | $KH_2PO_4$                           | 1 g l <sup>-1</sup>     |                                    |  |
|                      | MgSO <sub>4</sub> .7H <sub>2</sub> O | 0.5 g l <sup>-1</sup>   |                                    |  |
|                      | NaCl                                 | $0.1 \text{ g } 1^{-1}$ |                                    |  |
|                      | CaCl <sub>2</sub>                    | $0.1 \text{ g } 1^{-1}$ |                                    |  |
|                      | pН                                   | 5.5                     |                                    |  |
|                      | Incubation period                    | 120 h                   |                                    |  |
| PHM                  | Sucrose                              | 10 g l <sup>-1</sup>    | $6.12 \pm 0.11$                    | $29.01 \pm 0.07$                               |
| (after optimization) | Yeast extract                        | 5 g l <sup>-1</sup>     |                                    |  |
|                      | $KH_2PO_4$                           | 1 g l <sup>-1</sup>     |                                    |  |
|                      | MgSO <sub>4</sub> .7H <sub>2</sub> O | 0.5 g l <sup>-1</sup>   |                                    |  |
|                      | NaCl                                 | $0.1 \text{ g } 1^{-1}$ |                                    |  |
|                      | CaCl <sub>2</sub>                    | $0.1 \text{ g l}^{-1}$  |                                    |  |
|                      | pН                                   | 5                       |                                    |  |
|                      | Incubation period                    | 120 h                   |                                    |  |

Basal Media components were dissolved in 1000 ml of distilled water. Peel Hydrolysate Media components were dissolved in 1000 ml of PPH (pH 5.5) inoculated with 1% inoculum followed by incubating for 120 h at 25°C. The biomass (dry weight) and protein content (percentage of protein in dried biomass) data represents the mean of triplicates ± standard deviation.

# 3.3. Bioprocess optimization for enhancing SCP production

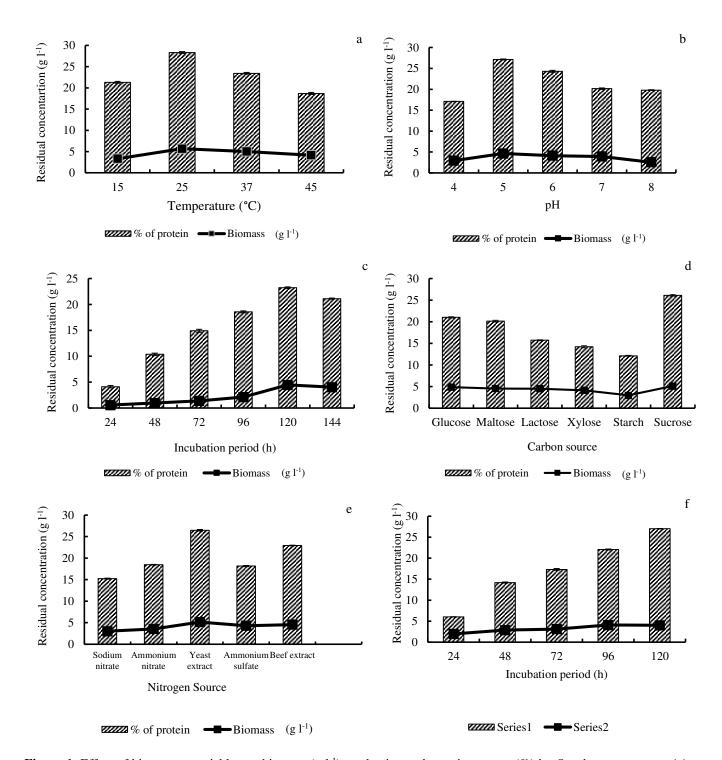
Incubation temperature includes an obligate correlation with the life process of every microorganism used in fermentation [36]. Every microorganism includes an optimum temperature level, at which they exhibit maximum growth and production yield. Temperature variation affects chemical/enzymatic compositions, nutrient requirements and growth rates of the microorganisms [37,38]. Therefore, temperature plays a vital role, which must precisely be monitored in every fermentation process. Effects of various incubation temperatures on SCP yield were assessed for a time period of five days. In the present study, the highest protein content (28.32%), corresponding to biomass content (5.67 g l<sup>-1</sup>), was achieved when the production media was incubated at 25°C (Figure 1a). Result of the present study was similar to result of the study by Ritchie et al. [39]. They reported that the optimum mycelia growth in Rhizoctonia solani isolated from potato occurred at 25°C (pH 5.6) when cultivated on potato dextrose agar. Similar results were reported for SCP production using papaya wastes as the substrates; in which, a maximum biomass of 4.67 g l<sup>-1</sup> was achieved at 25°C (pH 5) using submerged fermentation [22]. The pH of the fermentation media has great effects on protein production. Increases in pH beyond the optimum levels result in gradual decreases in protein contents due to the changes in solubility of media components and alterations in cell membrane permeability. Result of the current study revealed that the maximum protein content (26.03%) was achieved when the PPH was preserved at pH 5 (Figure 1b). These results were similar to previous results from effects of pH on microbial protein production [40-42].

The maximum production of SCP from Okara-wheat grit substrate was recorded at pH 5 (incubated at 25°C and 150 rpm) using *Rhizopus* (*R.*) oligosporus (protein contents of 35.98%) and *Aspergillus* (*A.*) oryzae (protein contents of 36.20%) using submerged fermentation [42].

In this study, effects of incubation time on SCP production were studied by inoculating the fermentation media with 1% of inoculum, followed by incubating at 25°C for various time periods. Optimization of the incubation time revealed that the maximum SCP production (4.86 gl<sup>-1</sup>) and protein content (23.23%) were achieved within 120 h of incubation (Figure 1c). In a previous study on orange peels as fermentation substrates for SCP production, the maximum protein content (31.12% w w-1) was observed within 120 h of incubation using R. oryzae [43]. The gradual decreases in protein and biomass contents after 120 h of incubation could be highlighted as significances in nutrient depletion and production of undesired waste debris by yeasts, as suggested by previous studies [43]. In the present study, findings of the biomass protein production from orange wastes using R. oryzae were similar to those reported by other studies. In fact, R. oryzae demonstrated a maximum protein content of 22.83% when cultivated in orange peel media (pH 5) at 25°C for 120 h at 120 rpm using submerged fermentation [43]. Screening for ideal carbon and nitrogen sources for enhanced SCP production includes as an important step in calculating overall costs of the production. Results in Figure 1d showed that the maximum protein production (26.13%), corresponding to elevated biomass level of 5.11 g l<sup>-1</sup>, was achieved using sucrose. Sucrose has been reported as an ideal candidate for SCP production by A. niger grown in orange peel media. Indeed, A. niger recorded a maximum protein content of 22.83% when

cultivated in orange peel media (pH 5) at 25°C for 120 h at 120 rpm using submerged fermentation [43]. Carbon plays a significant role in energy generation and product biosynthesis in yeasts and the preference of carbon source

varies depending on the type of yeasts used for fermentation [41,42].



**Figure 1.** Effect of bioprocess variables on biomass (g  $l^{-1}$ ) production and protein content (%) by *Saccharomyces cerevisiae* NCDC 364 (a) Effect of Temperature (b) Effect of pH (c) Effect of Incubation period (d) Effect of Carbon source (e) Effect of Nitrogen source (f) Increased biomass (g  $l^{-1}$ ) production and protein content (%) after optimization.

In yeasts, nitrogen source acts as an important agent in synthesis of the cell structural and functional nitrogenous components. Decreases in nitrogen source slow down growth in yeasts due to inhibition of transporting proteins that help sugar transports across the cell membrane [41]. In the present study, the maximum protein content (26.47%) was achieved using yeast extract as nitrogen source (Figure 1e). Beef extract was shown as an ideal nitrogen source for SCP production using papaya peel wastes with a yeast yielding total protein content of 29.73% (w w<sup>-1</sup>) (pH 5) at 25°C through submerged fermentation [22]. In this study, combined effects of the optimized parameters on overall increases in biomass yields and protein contents were investigated as well. Findings of the fermentation studies before and after optimization are demonstrated in Table 2. Moreover, increases in biomasses using optimized media are shown in Figure 1f. A maximum increase in protein contents of up to 29.01% was achieved under optimized conditions. In earlier studies using Candida utilis for biomass production in distillery wastes, protein contents of 46.12% were achieved under optimized temperature of 30°C and pH of 5 [42]. Protein contents of 51.2% have been reported in potato waste waters supplemented with 5% of glycerol using Candida utilis ATCC 9950 through submerged fermentation [43]. In other studies, including lemon and orange peels as substrates for SCP production, protein contents of 52.1 and 56.9% were respectively reported when Rhodococcus opacus DSM 1069 was used as fermenting microorganism [44]. Findings from the present study were similar to findings from previous studies on production of SCP from pineapple wastes using S. cerevisiae (28.6% w w-1 dry weight of SCP) and glucose (2%) as carbon sources [43]. Statistical optimization of bioprocess variables such as temperature (28°C), incubation time (120 h), seed size (10% v v<sup>-1</sup>), seed age (48 h) and concentration of date sugars (10 g l<sup>-1</sup>) has been reported to enhance the protein content up to 47.34% from Fusarium venenatum growth on modified Vogel media [45].

#### 3.4. Amino acid profiling of SCP

The AA profiling of SCP was carried out using HPTLC (Table 3). In general, 18 AAs detected in the samples; of which, ten are essentials and eight are non-essentials in

nature. Tryptophan was found in a comparatively higher concentration of 6.52%, followed by threonine (3.25%). Other EAAs were found in relatively lower concentrations. Non-EAAs of higher levels included glycine (5.76%) and glutamic acid (4.43%). Pineapple peel based SCP included 55.07% of the total AA content. This result is strongly supported by the results of the previous studies [41]. Lack of the self EAA synthesis is an innate mechanism of microorganisms; for which, supplementation via diets becomes mandatory. Appreciable levels of EAAs and non-EAAs in the produced SCP are linked to SCP proteinaceous nature, enabling its possible uses as a protein-rich ingredient/supplement in livestock/aquaculture feeds. However, further toxicological assessments and animal trials are necessary.

#### 4. Conclusion

In this study, rich nutritive profile of customarily discarded pineapple peels was investigated as cost-effective substrates for the production of commercially important SCP using S. cerevisiae NCDC 364. Steam treatment and acid hydrolysis methods were used as appropriate methods, compared to routinely used enzyme treatment or other chemical pretreatment methods. A sequential bioprocess optimization strategy was successful in enhancing SCP production using PPH. The present study highlights the importance of easily available, carelessly discarded waste raw materials such as fruit peels as potential substrates for the production of SCP through biotechnological methods. Furthermore, significant effects of bioprocess variables in SCP production were investigated. Although the optimized media recorded an increased biomass and protein content, use of statistical optimization tools such as RSM can further improve the production strategy, considering interaction effects of bioprocess variables. Moreover, replication of the optimized results in fermenters followed by scale-up studies can help to address problems associated with commercial SCP production using pineapple peels. In conclusion, results from this preliminary study can tackle global dilemmas of protein malnutrition when subjected to statistical optimization and scale-up studies.

Table 3. Amino acids profile (in percentage dry weight) of SCP extracted from optimized Peel Hydrolysate Media

| Essential amino acids (%)     |           |               |          |         |            |            |           |            |               |  |  |
|-------------------------------|-----------|---------------|----------|---------|------------|------------|-----------|------------|---------------|--|--|
| Leucine                       | Histidine | Methionine    | Arginine | Lysine  | Isoleucine | Tryptophan | Threonine | Valine     | Phenylalanine |  |  |
| 2.88±                         | 2.74±     | 3.23±         | 1.34±    | 2.55±   | 2.62±      | 6.52±      | 3.25±     | $3.09 \pm$ | 3.00±         |  |  |
| 0.014                         | 0.005     | 0.040         | 0.15     | 0.013   | 0.021      | 0.011      | 0.026     | 0.12       | 0.016         |  |  |
|                               |           |               |          |         |            |            |           |            |               |  |  |
| Non-essential amino acids (%) |           |               |          |         |            |            |           |            |               |  |  |
| Glutamine                     | Proline   | Aspartic acid | Alanine  | Glycine | Glutamic   | Asparagine | Cysteine  |            |               |  |  |
|                               |           | _             |          | -       | acid       |            |           |            |               |  |  |
| 2.52±                         | 2.09±     | 2.29±         | 1.10±    | 5.76±   | 4.43±      | 3.28±      | 2.38±     |            |               |  |  |
| 0.002                         | 0.007     | 0.013         | 0.003    | 0.011   | 0.030      | 0.018      | 0.005     |            |               |  |  |

The data represents the mean of triplicates  $\pm$  standard deviation.

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#### 6. Conflict of interest

The authors declare no conflict of interest.

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# افزودن ارزش پوست آناناس با تولید پروتئین تک یاخته با استفاده از *ساکارومیسس سرویزیه* NCDC 364

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### چكىدە

سابقه و هدف: پوست آناناس حاوی مقادیر قابل توجهی کربوهیدرات است که می تواند به عنوان ماده خام ارزان برای تولید محصولات مهم تجارتی از طریق تخمیر مورد استفاده قرار گیرد. هدف مطالعه حاضر استفاده از این ماده اولیه برای کشت ساکارومیسس سرویزیه NCDC 364 و کاربرد آن به عنوان پروتئین تک یاخته بود.

**مواد و روش ها:** پروتئین تک یاخته با استفاده از پو ست دور ریز آناناس و ساکارومیه سس سرویزیه NCDC 364 تولید شد. بهینه سازی متغیرهای زیست فرآوری (درجه حرارت، pH، مدت گرمخانه گذاری، منبع کربن، منبع نیتروژن) موثر بر تولید پروتئین تک یاخته با بررسی اثر یک عامل-در یک زمان انجام شد. یاخته های کشت داده شده در محیط کشت بهینه از نظر آمینو اسید به وسیله کروماتوگرافی لایه نازک با کارایی بالا غربالگری شدند.

یافته ها و نتیجه گیری: ساکارومی سس سرویزیه NCDC 364، در مقای سه با محیط بهینه ناشده، بی شترین میزان پروتئین تک یاخته از محیط بر پایه پوست آناناس را تولید کرد. بررسی اثر یک عامل - در یک زمان نشان داد بیشترین میزان تولید توده زیستی ابا مقادیر بهینه درجه حرارت ۲۵ درجه سلسیوس، pH برابر ۵، مدت گرمخانه گذاری ۱۲۰ ساعت، ۱ درصد ساکارز به عنوان منبع کربن و 0۰ درصد عصاره گاو به عنوان منبع نیترروژن به دست می آید. پروفایل آمینو اسید توده زیستی برداشت شده با کروماتوگرافی لایه نازک با کارایی بالا نشان داد میزان تریپتوفان موجود بیشتر از 00 در صد می با شد و بعد از آن ترئونین (00 (01 قرار دارد. نتایج این مطالعه پیشنهاد می کند که مواد اولیه ای که به راحتی قابل دسترس می باشند، مانند پوست میوه ها، رشد مایه هایی آمقرون به صرفه برای تولید تجاری پروتئین های میکروبی مهم به منظور رفع مشکلات و نگرانی جهانی مربوط به سوء تغذیه پروتئین محسوب می شوند.

تعارض منافع: نویسندگان اعلام می کنند که هیچ نوع تعارض منافعی مرتبط با انتشار این مقاله ندارند.

#### تاريخچه مقاله

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#### واژگان کلیدی

- پروفایل اسید آمینه
- ساكاروميسس سرويزيه
  - پروتئین تک یاخته
    - يوست آناناس

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