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Upgrading of Tomato (*Solanum lycopersicum*) Agroindustrial Wastes

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Abstract

Utilization of Tomato Agroindustrial Wastes (TAW) is of growing importance for its low cost and availability. This work aimed to add value to TAW from the point of pharmaceutical and food aspects, environmental safety, and energy security. Results indicated that TAW are not only source of lipids, proteins and inorganic minerals but are also source of lignocellulosic biomass for production of biofuel and single cell protein.

Keywords: Tomato agroindustrial wastes; Valuable products; Enzymatic hydrolysis; Bioethanol; Single cell protein

Introduction

The high production and disposal of agricultural wastes and their uncontrolled decomposition generate environmental pollution [1]. Worldwide research is in progress to minimize the negative effects of such pollutants in conjunction with the development of potential market demand of value added bio-commodities, such as; fermented beverages, Single-Cell Proteins (SCP), Single-Cell Oils (SCO), bio-colors, flavors, fragrances, polysaccharides, bio-pesticides, plant growth regulators, bioethanol, biogas and bio-hydrogen [2]. Tomato is one of the most significant vegetables' worldwide productions [3]. Egypt is the world fifth producer of tomato (*Solanum lycopersicum*), annually producing over than nine million tons [4]. Thus Egypt has huge amounts of tomato crop residues and agroindustrial wastes; leaves, stems, pomace (skin, seeds and pulps). Tomato Agroindustrial Wastes (TAW) could be used for producing different valuable products, such as carotenoids, proteins and it can be used also as a potential feedstock for ethanol production not only because of their low cost and high availability but also because of their considerable amount of sugars and low content of lignin [5].

This work aims to investigate the upgrading of TAW into high value extracts, bioethanol and single cell protein.

Materials and Methods

Media

For cultivation, maintenance and preparation of yeast for bioethanol fermentation, Wickersham's medium was used [6].

Waste samples and their constituents

Tomato crop residues; roots, stalks and leaves were obtained from agricultural field in El-Gharbia governorate, Egypt. While, the tomato processing residue, i.e., the tomato pomace was obtained from food industry (Durra food products) 10th Ramadan city, Ash Sharqiyah governorate, Egypt. The protein content was determined according to AOAC (1970), lignin content was determined By one, while the cellulose and hemi-cellulose contents were determined by some other researchers.

Pretreatment of tomato wastes

The Tomato Agroindustrial Wastes (TAW) were first defatted according to the method reported by Hussein et al. [7]. Then the defatted wastes were delignified by alcoholic sodium hydroxide solution according to the method reported by Zhao et al. [8].

Enzymatic saccharification and reducing sugar assay

Enzymatic saccharification of the pretreated TAW was carried out using partially purified cellulases and hemicellulases obtained using different fungal strains available in laboratory; endoglucanase CMCase 173.3 IU/g protein and exoglucanase FPase 17.33 IU/g protein from *Synchytrium endobioticum*, cellobiase 19.43 IU/g protein from *Aspergillus niger* and xylanase 106.9 IU/g protein from *Penicillium chrysogenum*. The enzyme mixture (cellulase+cellobiase) was applied at zero time, while hemicellulase was added after 24 h of starting the reaction. The hydrolytic reaction was carried out within 72 h, at 50°C using 0.05 M citrate buffer (pH 4.8).

Total Reducing Sugars (TRS) were determined by Nelson-Somogyi assay and TRS values were calculated as the equivalent of glucose. The saccharification percentage was calculated according to Abo-State et al. [6] and Gusakov et al. [9].

$$\text{Saccharification precentage} = \frac{\text{Formed TRS} \times 0.9}{\text{Cellulose content in pretreated substrate}} \times 100$$

Batch bioethanol fermentation

This was carried out according to Abo-State et al. [6] where peptone (10.0 g/L), KH_2PO_4 (2.0 g/L) and $MgSO_4.7H_2O$ (1.0 g/L) were added to the hydrolysate and then sterilized by autoclaving at 121°C for 20 min. The medium was inoculated with 10% (v/v) cell suspension of *Saccharomyces cervisiae* and then incubated at 30°C for 72 h at 150 rpm. Ethanol concentration was determined according to the method of Raid and Truelove and residual TRS was also determined. Then ethanol yield was calculated according to Abo-State et al. [6].

$$Ethanol\ yield = \frac{Produced\ ethanol}{Utilized\ TRS} \times 100$$

The efficiency of ethanol production was calculated according to Caylak and Sukan [10].

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$$Ethanol\ production\ effeciency(\%) = \left(\frac{Produced\ ethanol\ g}{Utilized\ TRS\ g}\right) \times \left(\frac{1}{0.511}\right) \times 100$$

where, 0.511 is the theoretical yield of ethanol produced from glucose.

All of experiments were carried out in duplicate and illustrated data are mean values of obtained results.

Results and Discussion

The constituents of different collected tomato wastes

The results listed in Table 1 revealed that the different collected TAW constituted of different valuable constituents. The lipid fraction, containing carotenoids, ranged between 0.55 and 4.1 wt%. The protein fraction ranged between 10.22 and 24 wt%, revealing that TAW is rich source of essential amino acids. The lignin content ranged between 6.1 and 10.5 wt%. While the holocellulose (cellulose and hemicellulose) represented the main content ranged between 63.92 and 74.9 wt%. But TAW contained lower content of inorganic minerals i.e. ash that ranged between 2 and 6 wt%. Thus, TAW is not only a green source of lipids and proteins with good nutritional quality but also a source of lignocellulosic matter with potential for bioethanol production [10].

Guuntekin et al. [11] reported that holocellulose is the major component of tomato stalks, counting 88%, with approximately 40.35% and 47.65% cellulose and hemicellulose, respectively, while its lignin content represents approximately 4.15%. Yargıç et al. [12] reported that cellulose and ash compromise about 34.59% and 4.49% of tomato wastes. Karthika Devi et al. [13] showed similar results in protein content of tomato peels and seeds, the main components of tomato pomace that counted approximately 16.19 and 26.39%, respectively, with ash content of approximately 2.50 and 3.5%, respectively. Achmon et al. [14] illustrated that tomato pomace contains 4.46% ash. Tomato peels are reported to contain, on dry basis, 14-20% protein, 40-74% lignocellulosic materials and 3-5% lipids. While, the tomato seeds are shown to contain 20-40% protein, 35-50% lignocellulosic materials and 18-37% lipids [15-17].

Pretreatment of tomato wastes

The data of Table 1 collectively indicated the presence of variable proportions of non-carbohydrate obstacles for bioconversion of TAW. This included lignin, lipids proteins and inorganic salts (appeared as ash). Thus pretreatment of these wastes, aiming at defatting followed by delignification was done. Pretreatment methods refer to the solubilization and separation of one or more of these components of biomass.

In this study, the pretreatment included physical pretreatment by grinding followed by alkaline chemical pretreatment. That led to approximately 50% reduction in lignin content of TAW. Where, the pretreated samples of tomato stalks, tomato leaves, tomato roots, tomato pomace comprised residual lignin of approximately 4.30, 3.40, 4.24, 3.60 wt%, respectively.

In lignocellulosic material, NaOH gives better internal surface by swelling it and leads to lignin degradation. NaOH pretreated lignocellulosic biomass results higher porosity that leads to better glucose yield after enzymatic hydrolysis by attacking the ester bonds.

Employing dilute NaOH is wiser than employing concentrated NaOH for the environmental and economic benefits [8,18].

Saccharification of pretreated tomato wastes by partially purified cellulases and hemicellulases

One of the most important targets (of the present work) is the saccharification of the pretreated samples of tomato wastes using partially purified cellulases and hemicellulases. The results illustrated in Figure 1. indicated that regardless of the type of TAW, the increase of reaction time, increased the saccharification percentage. That recorded; 42.62, 51.34 and 43.51% for tomato stalks, leaves and roots, respectively after 72 h. While, tomato pomace recorded the highest saccharification

Tomato crop residues	Cellulose (%)	Hemicellulose (%)	Lignin (%)	Lipid (%)	Protein (%)	Ash (%)
Stalks	22.12	41.80	7.85	1.56	24	2.67
Leaves	26.15	48.75	6.10	3.50	13.5	2.0
Roots	26.36	46.37	10.5	0.55	10.22	6.0
Pomace	38.01	31.42	6.87	4.10	16.6	3.0

Table 1: Chemical constituents of different residues of tomato agroindustrial waste TAW.

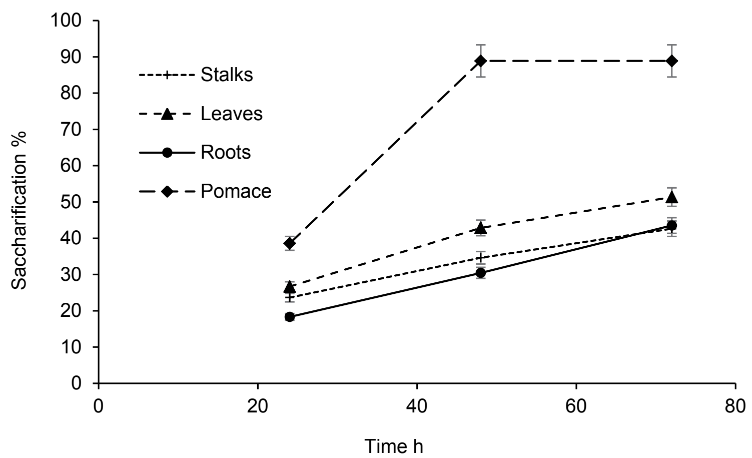

Figure 1: Saccharification of the pretreated tomato pomace by the partial purified cellulases and hemicellulose.

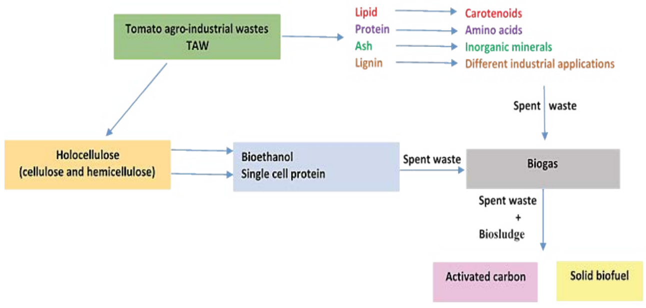

Figure 2: Valuable products from Tomato Agroindustrial Wastes TAW.

percentage of approximately 88.88% after 48 h, and remained sustained with longer incubation period. Accordingly, the hydrolysate of tomato pomace was taken for further bioethanol fermentation process.

Production of bioethanol and Single Cell Protein (SCP) from saccharified sample of tomato pomace

The final target of the present work was the production of bioethanol and microbial cells, as a source for SCP, from the most susceptible enzymatically saccharified TAW; the tomato pomace. With regards of the bioconversion of the hydrolysate to bioethanol by *S. cervisae*, it yielded 30.7 g ethanol/L hydrolysate of approximately 0.3 g bioethanol/g utilized TRS (i.e., bioethanol yield of ≈ 29.98%), with bioethanol fermentation efficiency of approximately 58.67%. Moreover, the batch fermentation process yielded 54.40 g SCP/L hydrolysate (Figure 2).

Conclusion

This study would provide an important reference for the concept and the feasibility of the production of valuable from tomato (*Solanum lycopersicum*) Agroindustrial Wastes (AW). Where, different sub-products and biofuels can be produced (Figure 2); lipids, protein and ash as reach sources of essential carotenoids, amino acids and inorganic minerals, respectively; lignin which has different industrial applications, holocellulose (cellulose and hemicellulose) for production of bioethanol and single cell protein and the spent wastes obtained from all the aforementioned extraction and biorefining processes can be used for biogas production. Moreover, to reach for the zero waste point; the spent wastes and biosludge after biogas fermentation, can be used for production of solid biofuel or activated carbon.

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