

An overview on the applications of solid-state fermentation in sustainable production of value-added products

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Abstract

The solid-state fermentation (SSF) is one of the oldest bioprocess which has been applied by humans for various food applications since ancient times. Though traditionally SSF was used for production of fermented foods viz. tempeh, koji, cheese, pickles and sausages, it has continued to draw attention for its utility in various sectors encompassing industrial, textile, pharmaceutical, bioenergy, food and environmental domains. The reason for continued interest in SSF can be credited to the simplicity of bioprocess, which involves growth of bacterial or fungal microorganisms on moist solid substrates, in absence or very scanty water. The use of various agro-industrial wastes as substrates in SSF, offering dual advantage of valorisation to high-value products with simultaneous waste management, adds to the rationale of SSF deployment for sustainable development with environmental benefits. The current review carries out critical analysis and summarizes the recent developments in application of SSF in various processes of commercial significance viz. production of enzymes, platform chemicals, aroma compounds, biopesticides and biological detoxification.

Keywords: Solid-state fermentation, agro-industrial wastes, biomass, enzymes, platform chemicals, biological detoxification

Introduction

Solid-state fermentation (SSF) is a bioprocess which is carried out in near absence of water by cultivating microorganisms on moist solid-substrates. These inert supports act as energy or carbon source for cultivated bacterial or fungal species and help in their growth with simultaneous production of useful metabolites, enzymes by them. Thus, many versatile substrates possessing critical nutrient and moisture content, which can support the growth of the fermenting microorganisms can be valorised using SSF [1, 2]. These substrates include a plethora of biomass ranging from food or kitchen waste residues, forestry residues, agro-residues, industrial by-products which are either toxic or underutilized. Therefore, SSF has established itself as an effective bioprocess for nutritional enrichment of agro-residues by biotransformation, bioremediation or biodegradation of toxic compounds leading to biological detoxification of hazardous agro-industrial residues [3, 4].

Apart from availability and cost playing a critical role in selection of a residue as a substrate in SSF, other important aspects such as surface area, nutritional accessibility, crystallinity, particle size and porosity are also considered [5]. After the choice of substrate, the selection of appropriate fermenting microorganisms is one of the most critical aspects for effective SSF. Filamentous fungi have been considered as most attractive microorganisms for SSF with *Aspergillus oryzae* and *Penicillium roquefortii* being classical examples for production of koji and cheese respectively by SSF since ancient times. This could be attributed to their ability to grow in low water environment as well production of complex hydrolytic enzymes which enables them to effectively draw nutrition from recalcitrant lignocellulosic biomass [6, 7]. Nevertheless, other microorganisms viz. various strains of yeast, actinomycetes and bacteria have also been reported to be successfully used in SSF for production of various high value microbial products which include enzymes, antibiotics, organic acids, platform chemicals, biosurfactants, biopesticides etc.[1, 2]. Since SSF provides an environment which is close to natural habitat of microorganisms, this technique has enormous potential to convert a myriad of microorganisms into microbial cell factors for production of desired value-added products at commercial level.

Considering above perspectives, it can be well established that SSF offers several advantages for production of microbial metabolites with improved economic feasibility in sustainable and environmental friendly bioprocesses. The benefits offered by SSF include effective bioprocess for solid-waste management by valorisation of various agro-industrial residues, economic credibility as the utilized substrates bear no or very low cost, production of products in high titres, lower energy demand, minimized risk of contamination and simple downstream processing with minimal or no use of toxic organic solvents [7, 8]. Due to these advantages, SSF has been used for production of enzymes, fermented foods, animal feed at commercial scale worldwide, with it being most popular in Asian countries like Japan, China, Korea and India [1, 5].

These scaled up SSF processes are majorly based on use of bioreactors of different types viz. fixed bed, packed bioreactors, Raimbault columns, perforated trays, agitated bioreactors, horizontal drum, rotating drum bioreactors etc. depending on the production of desirable end products by appropriately chosen microorganisms and substrates [5, 7]. Despite these advantages, there are hindrances in easy scale up of SSF processes which include

difficulty in managing heterogeneity of the system, temperature control issues due to intense heat generation along with ease of instrumentation in its competitive counterpart i.e. submerged fermentation. Among one of the major drawbacks, the limited heat transfer capacity of the inert supports used in SSF leads to poor heat transfer which leads to denaturation of heat sensitive bio-products such as enzymes, antibiotics, aroma compounds as well as is detrimental to fermenting microorganisms [1, 5]. The use of thermophilic fermenting microorganisms producing robust and thermostable products has been advocated as an effective strategy to combat this limitation of heat sensitivity. The production of thermostable and ionic liquid stable xylanases with effective applications in xylooligosaccharide production, biomass saccharification has been reported by SSF of toxic *Jatropha curcas* seed cake by a thermophilic fungus i.e. *Sporotrichum thermophile* [9, 10]. Similarly, production of a thermostable β -glucosidase by *Thermomucor indicae-seudaticae*, a thermophile was reported by SSF of wheat bran [11].

Summarily, it can be stated that SSF is a highly efficient technique for three main reasons: (i) valorisation of abundant and underutilized or toxic residues (ii) economic and sustainable production of value-added products, as an alternate to adverse chemical synthetic pathways (iii) transformation of spent residue into nutritionally enriched substrate which can be used as feed or fertilizer. The present review highlights the potential of SSF as sustainable and green process for bioconversion of various substrates and summarizes its diverse applications. The use of SSF in production of value-added products viz. enzymes, platform chemicals, aroma compounds, biopesticides as well as in biological detoxification has been discussed in subsequent sections.

Production of enzymes by SSF

Enzymes have received a lot of attention, as biocatalysts are foreseen as environment friendly alternates to harsh chemical catalysts, carrying out non-renewable fossil based synthetic transformations. Though their industrial production has been reported since 1874, when rennin obtained from calves' stomach was used for manufacture of cheese by Christian Hansen, their production demand continues to rise as they find myriad of applications in various industrial processes [12]. The value of industrial enzymes market is projected to reach 8.5 billion US\$ in 2022 from the worth of 6.1 billion US\$ in 2017 [13].

To meet this high demand with commercial viability, their economical production is utmost concern and SSF is a very attractive choice in this regard. Since SSF utilizes waste substrates that bear negligible cost, the enzyme production costs are significantly lowered with dual advantage of valorisation of wastes, which otherwise being left unattended were contributing to environmental and economic concerns. Various agro-industrial residues viz. soybean cake, cottonseed cake, rapeseed cake, groundnut oil cake, jatropha seed cake, citrus peels, coffee husk, sugarcane bagasse, wheat straw, rice husks, wheat bran, rice bran, cassava bagasse have been effectively used as substrates for cost-effective production of plethora of enzymes such as lipase, phytase, cellulase, chitinase, xylanase, laccase, amylase, tannase, protease etc. by SSF [1, 2, 8].

Further, the titre of enzyme production is also high in SSF viz. 1.95 and 7.13 times higher production level of cellulase and pectinase was achieved by SSF by *Aspergillus niger*

NCIM 548 as compared to submerged fermentation [14]. The operativity of SSF under low water conditions minimizes the risk of contamination and thus, reduced demand for stringent sterile conditions in SSF further reduces the cost. The high titre (120,000 UA/g) lipase production by SSF was achieved under non-sterile conditions by microbial consortium isolated from wastewater sludges [15]. The highly cost effective crude enzymes viz. cellulases, xylanases, proteases, keratinases lipases produced by SSF of various agro-industrial wastes find intensive applications in biorefinery for biomass saccharification, biodiesel and bioenergy production, leather and tanning industries as well as detergent formulations [1, 16, 17]. The effective usage of these economically produced crude enzymes drastically reduces the industrial operational costs as the purification costs and labour for getting pure enzymes are cut down.

Production of platform chemicals by SSF

To face the energy crisis due to depleting fossil reserves, utilization of renewable biomass as sustainable feedstock for production of petro-derived chemicals has been an emerging research area. These compounds (composed of 2–6 carbons) can serve as base for production of plethora of industrially important chemicals and thus, these are referred as platform chemicals viz. 3-hydroxy propionic acid, lactic acid, succinic acid, γ -Aminobutyric acid (GABA) etc. Due to the immense applications of these platform chemicals in various sectors such as pharmaceuticals, cosmetic, food, textile, polymer, solvent industries, their bio-based production has received great attention [18-20]. The SSF bioprocess has been found to be instrumental in bio-based production of these platform chemicals and thus, utilisation of abundant biomass by SSF for valuable transformations seems a desirable pursuit. The market demand of GABA, a neurotransmitter with many important physiological functions has been on surge due its role as C4 platform chemical for production of bioplastics, valuable chemicals such as 2-pyrrolidone, polyvinyl pyrrolidone and nylon 4, which otherwise were petro-derived. Grewal and Khare [21], reported the cost effective production of GABA by SSF of toxic cottonseed cake and showed the application of microbially synthesized GABA in production of 2-pyrrolidone, a high-value petro-chemical. The production of another C3 platform chemical i.e. lactic acid with numerous applications in food, dairy, pharmaceuticals, textile and bioplastics has been effectively reported by SSF. Various residues such as cassava peel, wheat bran, food waste, tea waste, rice bran, sugarcane bagasse, wheat straw have been used as substrates in SSF for cost effective production of lactic acid [1, 22, 23]. Similarly, SSF have also been reported to be an important bioprocess for production of other platform chemicals viz. succinic acid, itaconic acid, 3-hydroxy propionic acid etc.[1].

Production of biopesticides by SSF

Though use of biopesticides as an alternate to conventional chemical pesticides has been widely advocated due to environmental concerns and adverse health effects of chemical pesticides, their commercial application is still limited due to high cost of their production. The use of SSF technology for cost-effective biopesticide production has been successfully demonstrated as many studies have reported more resistant and stable spores produced by SSF as compared to those produced by submerged fermentation [24, 25]. Recently, Sakdapetsiri et al. [26], showed that coconut husk and rice bran used as substrate in SSF led to effective production of β -1,3-glucanase and *Streptomyces similanensis* viable cells which acted as

biopesticide and protected against black rot disease caused by *Phytophthora palmivora* in orchids. The production of *Bacillus thuringiensis* derived biopesticide was reported under SSF conditions by using wastewater sludge, a negative-cost substrate [27]. Similarly, production of biopesticides by *Coniothyrium minitans* under SSF conditions has been extensively studied due to its viable application in protection of many important crops [28].

Production of aroma compounds by SSF

Aroma compounds producing an olfactory stimulus find extensive applications in pharmaceutical, cosmetic, food and chemical industries, due to which their market continues to grow. Since SSF simulates natural habitat of microorganisms, it has been found to be an effective method for their production, yielding higher titre as compared to submerged fermentation. Recently, Hamrouni et al. [29] reported that the production of an unsaturated D-lactone i.e. 6-pentyl- α -pyrone (6-PP) with a coconut-like smell, by *Trichoderma* sp. growing on sugarcane bagasse was 80 times higher in SSF as compared to submerged fermentation. Similarly, *Trichoderma asperellum* also produced high titre (7.36 mg/g) of 6-PP along with amylases and lipases, utilizing mixture of agro-waste substrates in scaled up studies using SSF bioreactor [30]. The production of many γ -decalactones, aroma compounds was achieved by *Yarrowia lipolytica* in SSF reactors [31]. The utilization of spent leaves of the *Eucalyptus cinerea* waste by *Pleurotus ostreatus* and *Favolus tenuiculus* under SSF conditions led to production of important aroma compounds i.e. 1,3,3-trimethyl-2-oxabicyclo[2.2.2]octan-6-one and 1,3,3-trimethyl-2-oxabicyclo[2.2.2]octan-6-ol with effective applications in pharmaceutical and food industries [32].

Environmental applications of SSF in biological detoxification of agro-industrial wastes

The wastes generated from agricultural and industrial sector are either burnt in open, dumped in landfills or discharged as effluents because of which they exert detrimental effects on environment. Many of these wastes contain anti-physiological, toxic anti-nutritional compounds viz. tannins, hydrogen cyanide, gossypol which restrict their utilization and add to safe disposal issues. In this context, SSF has proved to be an effective approach for biodegradation of toxic compounds present in agro-industrial wastes. The degradation of toxic compounds could occur due to enzymes or other metabolites released as part of normal metabolism by microorganisms during growth under SSF conditions [3, 7, 16]. In environmental context, SSF has emerged as an effective waste-management strategy by valorization of toxic agro-industrial residues into value-added products with concomitant reduction in their toxin level, making them suitable for utilization as feed or fertilizer. The release of enzymes or other metabolites by fermenting microorganism may not only degrade toxin but also lead to improved digestibility and enhanced nutritional quality [3, 4, 7]. Though many agro-industrial residues have been detoxified by SSF approach, but some of the notable examples are removal of toxin ricin in castor bean cake [33], phorbol esters in jatropha seed cake [34], glucosinolates in rapeseed meal [35], gossypol in cottonseed cake [16], caffeine in coffee husk [36], saponins in tea seed meal [37] and β -ODAP (3-N-oxalyl-L-2, 3-diaminopropanoic acid) in grass pea [38]. The biological detoxification by SSF has many advantages over physical and chemical techniques conventionally used for treatment of these residues such as better specificity, no adverse effects on other nutrients, eco-friendly and leads to simultaneous production of value-added products.

Conclusions

Though SSF is a traditional bioprocess, it continues to evoke interest in researchers due to the numerous advantages offered by it. The most notable among these is the pivotal role of SSF in waste to wealth strategy, which relies on biotransformation of waste residues into high-value products, which otherwise were adding to environmental and economic burdens. Thus, SSF has proved to be cost effective, sustainable and green approach for production of value-added products from wastes and contributing to circular bioeconomy. The SSF finds effective applications in production of plethora of value-added products viz. enzymes, platform chemicals, biofuels, antibiotics, aroma compounds and biopesticides. Further, the role of SSF as a promising approach in biological detoxification of agro-industrial residues has added to its environmental applications. However, there are many challenges associated with the scale-up and operativity of SSF processes at commercial scale, which need to be worked on for optimally harvesting the true potential of this environment friendly and cost-effective bioprocess.

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