

Effects of Medium Formulation and Culture Conditions on Microbial Xylanase Production Using Agricultural Extracts in Submerged Fermentation (SmF) and Solid State Fermentation (SsF): A Review

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Abstract

There are several crucial fermentation parameters including carbon source, temperature, pH medium and agitation speed used to elucidate and optimise the production of xylanase in submerged fermentation (SmF) and solid state fermentation (SsF). These parameters are determined by the types of microorganism that yield xylanase. Precisely, suitable parameters allow the proper proliferation of microorganisms to produce high concentration of xylanase. Carbon source provides prerequisite nutrients for growth of microorganisms in SmF and SsF. In order to enhance the xylanase production, cheap but effective carbon source such as agricultural extracts is generally added to supply as the prime nutrient. SsF has become a known interest to produce xylanase because of its economical process of using agricultural extracts. Thus, lower cost production of xylanase is easily achieved. Therefore, proper optimisation of fermentation parameters would able to produce the desirable xylanase at the maximum level. Additionally, optimum pH medium and temperature possess huge positive impact on the growth of microorganisms and xylanase production. Indeed, optimal pH and temperature in agitated culture show greater xylanase activity compared to non-agitated culture. Nonetheless, further increase of the optimum agitation speed would cause irregular morphology of microorganisms that ultimately lead to xylanase interference. In the present day, the demand for xylanase is rising because of its remarkable usages in vast industries. The foremost industrial application of xylanase is involved in chlorine-free bleaching process in pulp and paper industry whereby xylanase is added into the pulp to degrade xylan found within the lignin residuals. Increasing applications of xylanase in various industries have developed xylanase to become more valuable. As a result, there are continuously endeavors to transform xylanase industry into a profitable market with lower costs of production using different types of agricultural extracts at the optimal growth conditions. All these aspects are reviewed in this article.

Statement of the Problem

Only a few studies on the optimisation of medium formulation for the maximum microbial xylanase production have been conducted. The application of agricultural wastes as the carbon source for the industrial xylanase production are scarce and not comprehensively studied, compared and reported in submerged fermentation (SmF) and solid state fermentation (SsF). Being the simple, non-toxic and cost-effective carbon source to yield xylanase, the replacement of xylan as the substrate with agricultural extracts in SmF and SsF is of great interest particularly in industrial production. Agricultural extracts are the good alternative carbon source due to their similarity in the polymers structure of xylan, as a result, these lignocellulose residuals are suitable to use as the prime carbon source for xylanase production. Besides that, the lack of precise information of the optimum growth conditions on the microbial xylanase production in SmF and SsF also lead to the vast studies over the past few years.

Keywords: Xylanase production; Agricultural extracts; Carbon source; Culture conditions; Submerged fermentation (SmF); Solid state fermentation (SsF)

Introduction

Xylanase is inducible enzyme which responsible for the complete hydrolysis of xylan into simpler compounds, mainly xylose [1].

Xylanase is genetically single chain glycoproteins with molecular weight of 6 to 80 kDa. Xylanase is active between pH 4.5 to 6.5 at 40 to 60°C. Xylanase is produced by numerous numbers of different fungi. Various strains of filamentous fungi such as *Aspergillus niger*, *Aspergillus oryzae* and *Trichoderma spp* have been reported to be the potent producers of xylanase. However, xylanase production is typically restricted to *Aspergillus spp* and *Trichoderma spp* in the industrial scale [2]. Meanwhile, *Aspergillus spp* are normally selected and optimised for xylanase production. Apart from xylanase, *Aspergillus spp* also produce huge variety of extracellular enzymes including amylase, cellulase and protease [3]. Xylanase shows tremendous potential in many industrial processes especially in textiles, leather, detergents and baking [4]. Both solid state fermentation (SsF) and submerged fermentation (SmF) are used in the production of fungal xylanase. SsF has become a popular approach to produce xylanase due to its economical value that does not involved complicated technology. Viniegra-Gonzalez et al. [5] studied the comparison between SsF and SmF in terms of enzymes production. In fact, they identified out that higher biomass and lower protein breakdown were among the factors involved in determining the production of enzymes in SsF. Besides that, the utilisation of inexpensive agricultural extracts in SsF is environmentally sound because besides providing sufficient nutrients as carbon source, it reduces pollutions to the surroundings. Hence, SsF is more economical compared to SmF. Indeed, SsF is an attractive and

economical method for xylanase production, especially for fungal cultivations. It produces higher enzymes productivity using lower operation and capital cost [6]. Malaysia with abundant natural rainforest will be of great advantage in xylanase production using SmF and SsF. Natural resources and agricultural extracts which are abundantly available in the environment such as rice bran, wheat bran, palm kernel cake and soybean hulls are used as potential carbon sources in xylanase production via SmF and SsF [7]. Nevertheless, continuous research and development efforts are being given to SsF to be as compatible as SmF, making it more practicable especially for industrial production. All in all, xylanase is one of the valuable enzymes which show immense potential in both biotechnological and industrial applications. Notably, filamentous fungi of *Aspergillus spp* have always been the preference choice because they produce higher activity of xylanase than other fungi, yeast and bacteria. In the present day, the demand for xylanase is increasing because of its prodigious utilisation in vast industries. Recently, there has been much concern and immense importance on xylanase due to its broad potential industrial applications especially pulp and paper industry. It is one of the leading industrial applications of xylanase whereby this enzyme is added into the pulp to degrade xylan found within the lignin residuals. As a result, it allows easy bleaching process of the pulp and hence, it increases the brightness of paper. Notably, xylanase plays an effective pivotal role in pulp and paper industry due to its simplicity and economical properties. Other applications of xylanase are in bakery, food and beverage, animal feed industry and so forth. There are always continuously endeavors to enhance xylanase into a profitable enzyme with lower costs of production, recovery and purification.

Xylan substrate for xylanase activity

Xylan or known as wood gum is a type of heterogeneous polysaccharide found in the cell wall of many plant species [8]. Xylan is consisted of complicated structures of hemicelluloses with the backbone of β -1,4-linked xylopyranose subunits with its substituent of O-acetyl, α -L-arabinofuranosyl, α -1,2-glucuronic or 4-O-methylglucuronic acid which are accounted for approximately 15 to 30% of the total dry weight in wood component of angiosperms [9]. Hemicellulose is also called as heteropolysaccharides comprised of low molecular weight polymers that consisted of 80 to 200 units degree of polymerisation [10]. Thus, it is also collectively viewed as the second most abundant biomass component in plants besides cellulose [11]. Hemicellulose has broad applications because of its non-toxic biodegradable features. Apart from that, hemicellulose is bonded to other components such as cellulose, proteins, lignin and phenolic compounds by hydrogen, covalent, ionic bond as well as hydrophobic interaction [12]. Xylan provides structural support to the plants besides polysaccharides storage in the seeds for germination purposes. Xylan of the plant cell wall is greatly differed depending on their origins and molecules that attached to the xylan backbone. The degradation of xylan occurs when β -1,4-xylanase cleaves the polysaccharide backbone of xylan followed by β -xylosidases hydrolyzes xylo-oligosaccharides to xylose [13]. Xylan is categorised into homoxylans, glucuronoxylans, arabinoglucuronoxylans, arabinoxylans, glucuronoarabinoxylans and heteroxylans depending on its degree of substitution and classes of side groups [14]. Homoxylans are typically found in seaweeds, glucuronoxylans are found in hardwoods and arabinoglucuronoxylans are found in softwoods. Hardwood xylan likes O-acetyl-4-O-methylglucuronoxylans has higher degree of polymerization about 150-200 than softwood xylan, arabino-4-O-methylglucuronoxylans

which only about 70-130 [15]. On the other hand, arabinoxylans, glucuronoarabinoxylans and heteroxylans are particularly found in cereals. In addition, cereal xylan consists of D-glucuronic acid [15]. The naturally occurring lignocellulosic plant biomass such as sawdust, sugarcane bagasse, barley, wheat bran and rice bran are mainly consisted of three groups of polymers, which are cellulose, hemicellulose and lignin. Cellulose and hemicellulose are sugar rich raw materials which often used in fermentation process. Hemicellulose is a linear and heterogenous mixture of five different sugars consists of D-xylose, D-mannose, D-glucose, D-galactose and L-arabinose. Hemicellulose has shorter chains and its amorphous, not crystalline structure made it easier to hydrolyze than cellulose [16].

In general, xylan is a yellow, water soluble, and gummy polysaccharide which is found in most of the plant cell wall and hydrolysed to produce xylose. Hydrolysis of xylan is an important step to liberate valuable products, mainly xylose. Xylose can be further process into ethanol. Hydrolysis of xylan can be done by two different methods which are chemical hydrolysis and enzyme hydrolysis. Chemical hydrolysis of xylan is typically used in many industries but results in number of toxic and undesirable substances that are being produced during the process causes the pollution to the environment. In order to get rid of the problem, enzyme hydrolysis of xylan such as xylanase to hydrolyse xylan into xylose which is environmental friendly is more preferable [17]. Xylitol, a five-carbon sugar alcohol is one of the major end products of xylan and is produced by fermentation of xylose. Xylitol has been widely used as a natural food sweetener, sugar substitute for diabetics and also as a dental caries reducer [16]. In the past, the conversion of xylan into xylose was conventionally carried out using acid hydrolysis process. Nowadays, many studies considered xylan as one of the new substrates for the production of biofuels, pharmaceuticals and solvents.

Xylanase

Xylanase or endo- β -1,4-xylanase is an extracellular enzyme that comprised of β -xylosidase, arabinofuranosidase and acetylxylanase [18]. These enzymes are found to be synthesised by eukaryotes and prokaryotes. However, the main producers are amongst fungi followed by bacteria. In general, xylanase has been categorized as glycosyl hydrolase Family F10 and G11 based on the amino acid sequence similarity and three-dimensional structure analysis. Predominantly, fungal xylanase is belonged to Family F10 because greater parts of its members are endo- β -xylanase which is larger with higher molecular mass approximately 35 kDa than Family G11 xylanase which is only about 20 kDa, hence they have better capability in cleaving the glycosidic bonds [19]. Torronen et al. [19] reviewed that the 3D structures of Family G11 xylanase has the overall shape of a "right hand". A wide variety of fungi and bacteria produce extracellular xylanase which act on hemicellulosic material to liberate xylose. The main highlight of xylanase is its efficiency in the degradation of xylan into several xylose units by cleaving β -1,4-glycosidic bonds of xylan backbone in a random manner within the lignocellulosic materials [13]. Additionally for a complete degradation of xylan, β -xylosidase proceeds cooperatively by hydrolysing the xylooligosaccharides into xylose. In this respect, the end product xylose are being utilised commercially in vast industries worldwide. Xylanase is the advantageous choice for the degradation of xylan because it has high specificity with slight reaction state, insignificant loss of substrate and side product output [15]. Studies have shown that the production of xylanase, an inducible enzyme is influenced by either pure xylan or substrates that contain generous amount of xylan.

Xylanase activity

Since xylanase is one of the most valuable enzymes in biotechnology applications, there are a lot of studies regarding xylanase activity. Xylanase activity is determined by measuring the reducing sugar released from xylan. In general, reducing sugar is the sugar that contains aldehyde group which is oxidised by oxidising agent to become carboxylic acid. Xylanase activity is influenced by the incubation temperature in the range of 45 to 60°C [20]. Furthermore, xylanase activity is also depended on the types of substrates. Other types of buffer used to dissolve substrate include sodium acetate buffer, pH 5.0 [21], phosphate buffer, pH 5.0 [22], citrate phosphate buffer, pH 6.0 [23] and potassium phosphate buffer, pH 6.0 [24]. In fact, there are several numbers of xylanase activity assays used for the determination of reducing sugars, each with their own definition of xylanase activity unit. These xylanase activity assays are differed in their assays procedure such as temperature, duration of incubation and substrate used. However, the principle is to quantify xylanase activity from the detection of reducing sugars released from the respective substrate. For example, one unit of xylanase activity is defined as the amount of xylanase required to release one micromole of xylose per mL of enzyme extract under the assay condition. Indeed, the detection of the reducing sugar xylose can be determined using several methods. One of the methods was from Khanna and Gauri [25] that required the addition of sodium phosphate buffer, pH 7.2 with incubation temperature at 37°C and incubation in a boiling water bath for 15 minutes. In a journal published by Gessesse and Gashe [26], xylanase activity was assayed with the aided of glycine-NaOH buffer, pH 9 and incubation at 50°C for 10 minutes followed by incubation for 5 minutes in boiling water. Another method was derived from Bailey et al. [27] in which citrate buffer, pH 5.3 with incubation temperature of 30°C and incubation in a boiling water bath for 5 minutes. Bailey et al. [27] reported that 3,5-dinitrosalicylic (DNS) method has been widely used in most laboratories as the most common and effective method in determination of xylanase activity [28]. DNS method uses xylose standard curve as a standard to determine the amount of reducing sugar released. In addition, Rochelle salt used in DNS method plays an important role in color stabilisation besides preventing the reagent from dissolving oxygen. The higher the amount of xylose, the darker the color of the enzyme-xylose complex formed. As a result, more light will be absorbed. The amount of reducing sugar released was quantified using DNS reagent by Miller [29]. The DNS reagent was used to halt the reaction between xylan and xylanase to produce xylose. Another method to measure the reducing sugar is the Nelson and Somogyi method. It requires incubation in a boiling water bath for 15 minutes. The addition of Somogyi copper reagent is used to stop the reaction process by reducing copper from cupric to cuprous and conclusively to cuprous oxide. With the addition of Nelson arsenomolybdate reagent, it aids in the reduction of molybdenum acid to molybdenum blue [30]. The measurement of the reducing sugar is conducted by spectrophotometric reading at 540 nm. Comparatively, DNS method is more common method to measure the reducing sugar of xylose compared to Somogyi and Nelson method because it is less hazardous, faster and more reliable [31]. DNS method was abundantly reported in most studies of xylanase activity assays to measure xylose according to Singh et al. [24]; Yang et al. [32]; Kavya and Padmavathi, [33]; Kaur et al. [34] and Murugan et al. [35].

Industrial applications of xylanase

Xylanase has become a growing interest lately because of their promising applications in various industries. Among those important xylanase industrial applications are pulp and paper [1,15,36] baking [1,37] and also food and beverage [1,7,33]. Xylanase is commercially applied to replace the use of harsh chemical such as chlorine which is known to be toxic and harmful to the environment during the bleaching process of kraft pulps in pulp and paper industry. Xylanase acts upon the surface layer of cellulose fibers to increase the permeability of the pulp for better chemical penetration through depolymerisation process [38]. Xylanase also acts within the inner fibers layer to aid in the bleaching process. As a result, xylanase pretreatment in pulp and paper industry has been reported to reduce the usage of chlorine. Utilisation of xylanase is more economical and environmental friendly, besides reduces the treatment time of pulps that results in greater final brightness of products [7,8]. In the countries likes Western European and North America, their environmental regulations are strictly restricted the use of chlorine in the bleaching process in pulp and paper industry [15]. Indeed, mannanase, a type of glycanase that helps in depolymerisation of hemicelluloses backbone of wood, especially on softwood, is combined with xylanase in order to improve the bleaching process of kraft pulp [15]. Consequently, xylanase as a low cost bio-bleaching agent gives benefit in both environmentally and economically.

Livestock feed such as corns and wheat that contains high amount of non-starch polysaccharide causes problem in nutrients digestibility due to the present of arabinoxylans that act as anti-nutritional factor. Consequently, xylanase supplement is added to improve effectiveness in nutrients digestion and absorption. In food and beverage industry, xylitol a product of xylose possesses wide utilisation in food, pharmaceutical, cosmetic and oral products. Xylitol is commonly produced as a sweetener for diabetics because it does not require insulin for its breakdown process. Additionally, xylitol also serves as sweetener to the glucose-6-phosphate dehydrogenase deficient community because it does not require glucose-6-phosphate dehydrogenase in its metabolism [39]. In addition, it demonstrates to provide good dental healthcare therefore xylitol is introduced into chewing gum, sweets, soft drinks, ice-cream and toothpaste. In general, xylitol is produced through two types of processes including chemical and microbiological. Chemical process involves the reduction of xylose from hemicellulose of xylan whereas another involves the hydrolysis of xylan into xylose followed by hydrogenation [40]. In the end, xylitol undergoes purification and crystallisation process [41]. Microbiological process involves the production of xylitol from xylose through hydrolysis of lignocellulose xylan-rich materials. Apart from that, xylanase in desirable amount also has been used to further fasten the bread making process to improve bread crumbliness, shelf life and bread volume [42]. In general, there are several of enzymes typically used in the baking process such as xylanase, protease, α -amylase, glucose oxidase, pentosanase and lipase. Enzymes used for baking are denatured to inactive protein during the baking process and thus they have to be thermo-labile enzymes. Hence, xylanase plays an important role in improving the water absorption in dough, increasing dough volume and decreasing dough firmness by changing the water insoluble hemicelluloses into soluble form. Moreover, xylanase also improves the gluten elasticity, bread crumb structure and volume by reacting to both soluble and insoluble pentosans in flour. Besides, shelf life of bread is prolonged by the addition of xylanase as antistaling agent in bread storage. Xylanase also help in improving the softening effect on dough [37]. Both Jiang et al.

[43] and Romanowska et al. [44] reviewed that xylanase from *A. niger* also aids in improving the loaf volume and bread quality.

In brewing industry, the problem of viscous polysaccharides such as xylan in the final beer filtration process is solved by pretreatment with xylanase to break down the xylan. As a result, it increases the filtration rates and prevents the accumulation of unwanted materials during the process. In juices making industry, enzymes are typically used in order to maximise the production of 'clear' juices. Xylanase is commonly added with other cell wall degrading enzymes such as pectinase and

cellulase in the clarification and filtration process to clarify the juice. In other words, xylanase plays a role in degrading pectins, starch and hemicellulose components to reduce turbidity and to increase the volume of juice during pulp processing steps [45]. Additionally, other potential applications of xylanase are involved in the bioremediation and bioconversion of agricultural, municipal and food wastes for the production of fermentable bioproducts and renewable biofuel such as bioethanol. Table 1 summarizes the major applications of xylanase in the industry.

Industry	Application	Reference
Pulp and paper Industry	As a bio-bleaching agent to improve the pre-bleaching process of kraft pulp	Wong et al. [8]; Kantelinen et al.[46]; Dietmar et al. [47]; Kulkarni et al. [15]; Pang and Omar [7]; Shah and Madamwar [36]; Gupta and Kar [1]; Kavya and Padmavathi [33]
Baking	Improve loaf volume, texture, stability and bread quality. Prolong the shelf life of bread	Bhat and Hazlewood [48]; Jiang et al.[43]; Kavya and Padmavathi [33]; Zulfiqar [37]
Food and beverage	Clarification of wine and juices. As alternative sweeteners and food additives	Dietmar et al. [47]; Pang and Omar [7]; Fang [49]
Feed industry	Improve animal feed digestibility and increase feed efficiency	Dietmar et al. [47]; Shah and Madamwar [36]; Maciel et al. [28]
Agriculture	Bioconversion of lignocelluloses into fuels and other chemicals	Gawande and Kamat [50]; Kavya and Padmavathi [33]

Table 1: Major applications of xylanase in industry.

Producers of xylanase

Recently, considerable attention has been focused on the use of microorganisms in industrial xylanase production. Microorganisms such as bacteria, fungi, yeast and actinomycetes are among the producers of xylanase. They are found to be active at temperature ranging from 40 to 60°C with the optimal pH medium in neutral scope for bacteria while slight acidic pH is the best condition for fungal xylanase. Nonetheless, there are microorganisms classified as thermophiles, alkaliphiles and acidophiles that are capable of producing xylanase. Thermophiles are microorganisms that are actively stable from 50 to 80°C. Thermophilic fungi such as *Paecilomyces thermiophila* J18 yielded 18580 U/g of xylanase activity within 7 days of fermentation by utilising wheat straw [51]. *Thermomyces lanuginosus* (D₂W₃) produced 48000 U/g of xylanase using sorghum straw within 6 days of fermentation [52]. Other types of thermophiles including *Caldicellulosiruptor spp*, *Thermoascus spp* and *Thermotoga spp*. The founding of thermostable xylanase enhances the production in pulp paper industry. Indeed, *Aspergillus spp* [53], *Penicillium spp* [54], *Streptomyces spp* [55], *Bacillus spp* [56] and *Trichoderma spp* [57,58] have been most comprehensively investigated and manipulated in the production of xylanase.

Alkaliphiles on the other hand are mostly from *Bacillus spp*. Microorganisms that fall into this category thrive well at pH above 8. A study by Kohli et al. [59] revealed that *Thermoactinomyces thalophilis* subgroup C produced xylanase actively at pH 8.5, *Streptomyces spp*. RCK-2010 at pH 8 [60] and *Bacillus spp*. Sam-3 at pH 8 [61]. Meanwhile, acidophiles is a term refers to microorganisms that are capable to flourish under pH 3. Acidophilic xylanase microorganisms are *Aureobasidium pululans* var. *Melanigenum* optimally produced xylanase at pH 2 [62] and *Penicillium occitanis* Pol6 at pH 3 [63]. The alkaliphilic xylanase application is important in pulp and paper industry whereas for acidophilic xylanase, it is needed

in food and feed industry. Kavya and Padmavathi [33] reviewed that xylanase production is higher in optimised conditions which reached 12.65 U/ml when compared to under optimised conditions which only about 9.38 U/ml. Majority of xylanase achieved their optimum xylanase activity in the range of pH 5.0 to 6.0, which are slight acidic condition at temperature between 40 to 70°C [64]. Meanwhile, Fang et al. [65] stated that both xylanase production and cell growth by *Aspergillus carneus* reached their optimisation at 30°C. On the other hand, fungi such as *Aspergillus spp* and *Trichoderma spp* are the main producers of xylanase as they have shown to produce the highest level of xylanase compared to yeast and bacteria [15]. Other reason would be fungi species are able to excrete out xylanase extracellularly. As a result, the extraction process of xylanase is conducted in ease. In fact, it is an economical way to produce this enzyme as fungi are cheaper to grow using widely available natural substrates rather than pure xylan. Table 2 illustrates the production of xylanase by various types of microorganisms.

Microorganism	Reference
<i>Aspergillus niger</i>	Pang and Omar [7]
<i>Aspergillus niger</i>	Ahmad et al. [66]
<i>Aspergillus niger</i>	Kavya and Padmavathi [33]
<i>Aspergillus carneus</i>	Fang et al. [65]
<i>Aspergillus foetidus</i>	Shah and Madamwar [36]
<i>Bacillus spp</i>	Gupta and Kar [1]
<i>Bacillus licheniformis</i>	Archana and Satyanarayana [67]
<i>Paecilomyces themophila</i>	Yang et al. [51]

<i>Penicillium chrysogenum</i>	Jayant et al. [68]
<i>Streptomyces spp</i>	Saurav and Kannabiran [69]
<i>Streptomyces cyaneus</i>	Ninawe and Kuhad [70]
<i>Trichoderma viride</i>	Juwaied et al. [71]

Table 2: Production of xylanase by different types of microorganisms

Production of xylanase via submerged fermentation (SmF) and solid state fermentation (SsF)

Multiple biotechnological approaches including SmF and SsF are utilised for xylanase biosynthesis. Using SmF, microorganisms are cultured in a liquid medium containing required concentrations of nutrients for the optimisation of medium formulation. Similarly, growth conditions of incubation temperature and medium pH are easily maintained in this fermentation process. In general, nutrients composition in SmF are supplied in the medium in the form of cheaper and readily available complex materials such as undefined agricultural wastes including rice bran and rice straw [72], corn cobs [73], sugarcane bagasse, oat straw and wheat bran [74]. SmF is now almost universal in the development of industrial enzymes in all other fermentation of industrial field. In the case of SmF, both of microorganism and substances are involved in submerged state in the liquid medium. Since the contents are in submerged state in the liquid medium, the modeling of the process is amenable and the transfer of heat and mass is more efficient [75]. Methods for the design of fermentation equipment and for the evaluations of its performance are greatly improved by increased knowledge of factors affecting oxygen transfer in those systems requiring some degree of aeration [76]. The production of commercially important enzymes in SmF has long been established.

Many reports have been studied on the xylanase production in the SmF and SsF [77-80]. Currently, 80-90% of xylanase are produced in submerged culture because of the microbial biomass and the substrates are homogeneously distributed in a liquid medium. Besides that, Gaanappriya et al. [81] also stated that SmF has a higher degree of intensification and higher level of automation. Due to the better understanding of scientific literature on the bacterial metabolism, characteristics and their response, it leads to the rising on the development of SmF for xylanase production. In order to develop a good fermentation, those parameters should be optimized and maintained according to the limitation of process, such as temperature, pH, substrate concentrations, size of inoculum, cultivation time and aeration. Sanghi et al. [77] has proposed their investigation on optimising the parameters on the production of xylanase from *B. subtilis* and its potential in bleaching of Kraft paper. Their work concluded that the optimisation of fermentation conditions enhanced xylanase production by 1.5 fold as compared to under optimised conditions. In SmF of *B. subtilis* for xylanase production may be initiated by direct inoculation of spores to the sterile liquid medium. Similarly, the medium is incubated at a specified temperature and pH with constant aeration and agitation until the biomass product is reduced. Irfan et al. [78] reported the selection of suitable medium also plays a vital role in xylanase production because it is a prerequisite to make the process cost effective. Besides that, in a study of Arthur and Elmer [76], they stated that aeration taken in the general sense to mean the provision of an adequate oxygen supply is required in some of degree for all aerobic

processes. When the transfer of oxygen supplied by aeration in SmF is treated as a series of rate processes, oxygen transfer equation for each step is developed. The physical absorption of oxygen is shown to be a function only of the design and operating characteristics of the equipment. Therefore, the advantage of using this technique for optimisation process is the ease of handling various parameters whereby they can be monitored, periodic sampling of broth, and controlled if necessary by the addition of further nutrients. According to Kavya and Padmavathi [33], low xylanase activity was obtained in non-agitated flasks, most probably due to oxygen or mass transfer limitations, while on agitation, high xylanase activity was produced, probably due to good oxygen supply. In addition, the volume used for fermentation also has a great impact on air supply, nutrient supply, growth of microorganism and production of enzyme [82,83]. Therefore, in SmF, aeration and agitation are very important to ensure availability of oxygen, nutrients and other essential substances to the growing cells. Table 3 shows previous reports studied on the production of xylanase by different microorganisms in SmF.

Microorganism	Xylanase (U/mL)	activity	Reference
<i>Bacillus circulans</i>	2.2		Mullai and Rene [84]
<i>Bacillus subtilis</i>	408.9		Irfan et al. [78]
<i>Bacillus subtilis</i>	695.12		Nagar et al. [85]
<i>Bacillus subtilis</i>	128		Annamalai et al. [86]
<i>Bacillus spp</i> 2129	19.1		Bocchini et al. [87]
<i>Streptomyces spp</i>	70		Nascimento et al. [88]
<i>Trichoderma viride</i>	2.453		Goyal et al. [89]
<i>Aspergillus niger</i>	44.1		Loera and Cordova [90]
<i>Aspergillus niger</i>	293.82		Bakri et al. [91]
<i>Mamillissphaeria spp</i>	242.7		Thanaporn et al. [92]
<i>Schizophyllum commune</i>	5.74		Haltrich et al. [13]
<i>Thermomyces lanuginosus</i>	2.70		Purkarthofer et al. [93]

Table 3: Previous reports studied on the production of xylanase by different microorganisms in submerged fermentation (SmF).

SmF is the culture of microorganisms in liquefied medium as compared to SsF which involves the growth of microorganisms in the absence or near-absence of free moving water. SsF has been defined as the growth of microorganisms on moist substrates under controlled condition in the absence of free-flowing water [36,94]. SsF is an effective biotechnological tool which shows greater potential for the production of large variety of enzymes. In addition, SsF is performed using different types of microorganisms, such as fungi, yeast and bacteria in the production of several enzymes including xylanase. Large numbers of bacterial and fungal species are known to grow well on limited moist substrates in the absence or near-absence of free water. SsF process generally uses raw natural material as carbon and energy sources. In other words, SsF is a cost effective and economical process due to the use of simple growth and agro-industrial residues as the carbon sources for the production of enzymes [95]. Agro-industrial residues or more commonly known as agricultural extracts from wheat bran, rice bran, soybean hulls and palm kernel cake are typically being considered as the best industrial substrates for SsF because they are abundantly available and low in cost [96]. In addition, SsF tends to generate value added products such as enzymes, flavors, organic acids, single cell protein and bioactive compounds from agro-

industrial residues [7,16]. Indeed, SsF possesses several advantages over SmF in the production of enzymes including xylanase. SsF is a simple process. It produces higher fermentation productivity with lower contamination risks. It uses little amount of water which exclusively reduces the wastewater output. Besides that, it involves less expensive downstream processing steps with lower capital costs and energy demand. The usage of cheap and abundant carbon sources especially agricultural residuals for SmF and SsF that often results in a higher productivity of enzymes [3,97]. In addition, SsF possesses lower risk of contamination because most of the contaminants including bacteria are not able to grow in the absence of free-flowing water [97]. Nevertheless, there are several setbacks of SsF such as temperature, pH, water content and diffusivity of substance across barriers during the growth of microorganisms. Heat is generated during the fermentation process, when the availability of water is meager. In SsF, heat is not able to regulate which eventually builds up and causes the enzyme denaturation [98]. Table 4 shows previous reports studied on production of xylanase by different microorganisms in SsF.

Microorganism	Xylanase activity	Reference
<i>Arthrobacter spp</i>	819 U/mL	Murugan et al. [35]
<i>Bacillus subtilis</i>	12.0 U/mL	Kim et al. [99]
<i>Bacillus subtilis</i>	8964 U/mL	Sanghi et al. [77]
<i>Bacillus subtilis</i>	5.19 U/mg	Heck et al. [100]
<i>Bacillus spp</i>	720 U/mL	Gessesse et al. [101]
<i>Bacillus pumilus</i>	142.0 U/mL	Kapilan et al. [102]
<i>Aspergillus niger</i>	23.97 U/mL	Pang and Ibrahim [103]
<i>Aspergillus niger</i>	12.65 U/mL	Kavya et al. [104]
<i>Coprinopsis cinerea</i>	695.8 U/mL	Kaur et al. [105]

Table 4: Previous reports studied on production of xylanase by different microorganisms in solid state fermentation (SsF).

Medium formulation for xylanase production

Medium composition is crucial to influence the xylanase activity. The production of microbial xylanase in SmF is strongly affected by the types of carbon source used. When Seyis and Aksoz, [58] studied the effect of different carbon sources on xylanase activity, sucrose showed higher xylanase biosynthesis by *Tetracladium marchalianum* compared to glucose in SmF. On the other hand, complex undefined agricultural extracts were also reported as more preferable carbon source for xylanase activity by several studies. Agricultural extracts or lignocellulosic biomass are important material resource and energy source which shows greater potential in lignocellulosic biotechnology application such as pulp and paper, food and feed, and bioethanol production [106]. As usual, agricultural extract constituents of cellulose, hemicelluloses, and lignin. Agricultural extracts contain high concentration of xylan about 15-30% in hemicellulose which is good as an inducer for xylanase production in SsF [107]. In addition, agricultural extracts such as wheat bran, rice bran, palm kernel cake, sugarcane baggase, maize, barley husk, soybean hulls and sawdust are typically used as carbon sources in the xylanase production of SsF [7,16,33,107]. All in all, lignocellulosic materials are utilised in culture medium formulation because they are plenteous in nature, low in cost and possess high level of carbohydrate content which are suitable to generate fermentable sugars. In some studies, nitrogen supplement of organic and inorganic sources were added to further increase the

growth of microorganisms and therefore enhanced the production of xylanase.

Effect of different carbon sources of agricultural extracts on xylanase production

Wheat bran

Endosperm cell wall of wheat bran or scientifically known as *Triticum aestivum* contains 75% of non-starch polysaccharide [108]. Wheat bran is a by-product of wheat milling industry in which it is originated from the outer layer of the wheat kernel. In general, wheat bran is defined as the outer hard layer of the grain. Wheat bran has been vastly used as animal feed for improvement of digestibility as well as in bakery products. According to Bergmans et al. [109], the vital components of wheat bran are consisted of 46% non-starch polysaccharides, 10-20% starch, 15-22% proteins and 4-8% lignin. Furthermore, arabinoxylan is the primary constituent of the non-starch polysaccharides in hemicelluloses. Wheat bran is considered as one of the extensively utilised carbon source for the growth of *Aspergillus spp* in the production of xylanase. Indeed, wheat bran is identified as the optimum carbon source for the production of xylanase by *Aspergillus spp*. According to Gwande and Kamat [50] and Kavya and Padmavathi [33], wheat bran was observed to produce the optimum xylanase activity of 9.87 U/ml by *A. niger* which was isolated from garden soil samples in Bangalore, India. They also observed that xylanase activity of 68.91 and 74.5 U/ml were produced by *A. terreus* 5 and *A. niger* 44, respectively. Wheat bran is abundantly available and highly nutritious agricultural by-product that commonly used in fermentation process. Moreover, wheat bran is an economical by-product which highly abounding with xylan, and thus, it possesses vital potential in xylanase production by microorganisms [50]. In a study conducted by Kavya and Padmavathi [33], they observed that wheat bran achieved the maximum xylanase activity of 9.87 U/ml in the xylanase production by *A. niger* in SsF. Much higher xylanase production of 74.5 U/ml was achieved using wheat bran by *A. niger* when compared to *A. terreus* [9]. In addition, Xu et al. [110] showed that xylanase production by *A. niger* XY-1 reached 14637 U/g dry substrate of wheat bran in shake flask under optimised condition.

Rice bran

Rice (*Oryza sativa*) bran is the outer layer of rice grain. It is a by-product of rice milling industry in polishing process which contains high in dietary fibers. It takes up 10% of weight in unpolished rice. In a study conducted by Fang et al. [65], rice bran contained high level of hemicellulose which was about 7.55 ± 0.49%. Hemicellulose of rice bran is a heteropolysaccharide which consists of a backbone xylan and sugars such as arabinose, galactose and glucose as side chains [111]. According to Kuhad et al. [106], they showed that xylan of rice bran contained high level of xylose which was about 46% xylose besides 44.9% arabinose. Hence, rice bran is an inexpensive and abundantly available carbon source which typically used in fermentation process [33,37,112]. A study was conducted by Kavya and Padmavathi [33], who observed rice bran was a good substrate on xylanase production by *A. niger* via SsF. Rice bran has been mainly supplied for animal feed as well as rice oil production in Asian counterparts. It has many nutritive values as unsaturated fatty acids with antioxidant and anti-cancer properties. However, the consumption of rice bran in human diet is limited due to the exposure of agro-toxics and contamination during the storage of rice bran [113]. On a global basis, over 600

million tons of rice was harvested annually due to its high amount of nutrition values [114]. Abundant agricultural extracts with estimated 4.5 million tons was produced annually worldwide mainly in China [115]. Rice bran has been suggested as the crucial carbon source for biotechnological production of enzymes including xylanase production by many different microorganisms such as *Streptomyces actuosus* A-151 [115], *Paenibacillus spp* strain HC1 [111] and *Arthrobacter spp*. MTCC 5214 [116], respectively.

Sugarcane bagasse

Sugarcane (*Saccharum officinarum*) bagasse is a by-product of sugarcane stalks in the form of fibrous residues after the production of sugarcane juice and sugar. Cellulose and hemicellulose are the two main polysaccharides that constituted 70% of the sugarcane bagasse [117]. Specifically, it contains 50% cellulose, 25% hemicellulose and 25% lignin [96]. This enormous amount of polysaccharides in sugarcane bagasse is useful for biofuels and alcohol manufacturing processes. Furthermore, it is being endeavored into animal feed, enzymes and organic acids production [118]. By utilising sugarcane bagasse, fungi such as *Penicillium janthinellum* that isolated from plant material found in a termite colony was able to produce xylanase activity of 98 U/ml [119]. Indeed, *A. niger* grown on Mendel and Weber media yielded 65.8 U/ml of xylanase compared to *A. niger* LPB 326 that produced 2099 IU/g of xylanase using 65% sugarcane bagasse and 35% soybean meal, respectively [28]. Notably, *Thermoascus aurantiacus* ATCC 204492 also produced 1597 U/g of xylanase using sugarcane bagasse as the sole carbon source [120].

Oil palm (Palm kernel cake)

Oil palm (*Elaeis guineensis*) is a tropical palm tree found abundant in Malaysia, Indonesia and Thailand. They are commercially cultivated for their edible palm oil which possesses characteristic as resistance to both oxidation and long term exposure to heat. Apart from that, palm oil has been utilised in cosmetic, detergents and biodiesel [121]. The rapid growth of this palm oil agricultural industry has developed Malaysia as the world's largest and most leading exporter of palm oil with the account of 52% of total world oils and fats exports in the year of 2006 [122]. Oil palm industry produces a lot of agricultural by-products, mainly palm kernel cake. Besides that, this oil palm waste also has a notable profitable market worldwide in which it is being used as animal feed because of its content that comprised of 15 to 18% crude protein. Again, Malaysia is the largest palm kernel cake exporter mainly to various Europe countries [123]. Due to its plentiful in the industry, palm kernel cake has been utilised as a potential substrate for the production of enzymes especially in SsF. It has been known that palm kernel cake was used in the cultivation of several fungi such as *A. niger* USM AI 1, *A. niger* II, *A. niger* F4, *Trichoderma spp* and *Phanerochaete chrysosporium* [124]. Indeed, palm kernel cake has also been applied as a substrate is more economical approach in the xylanase production using SsF due to its low cost value and enormous amount as agricultural by-product from oil palm industry [9,125]. In a study published by Phang and Ibrahim [126], *A. niger* USM AI 1 yielded 23.97 U/g of xylanase using palm kernel cake as the sole substrate. It was reported by Dusterhoft et al. [127] that palm kernel cake comprised of 6% xylan compound suitable for xylan production. Likewise, in a study carried out by Hakim [128], palm kernel cake produced the highest xylanase activity by *A. niger* using both SsF and SmF. Additionally, Pang and Omar [9] revealed that palm kernel cake contained high protein content which was a good substrate for xylanase production. From their results, palm

kernel cake was managed to achieve 9.5 U/g of xylanase production by *Aspergillus spp* through SsF. Furthermore, xylanase activity of 192.50 U/g was produced by *A. niger* FTCC 5003 in column bioreactor under optimised condition using palm kernel cake as the prime carbon source [129].

Soybean

Soybean (*Glycine max*) hulls are the outer shell of soybean seed that are being removed during the production of soy oil, soybean meal, protein and flour. They are commercially distributed in pellet forms as ruminant feed mainly for horses and cows. Soybean hulls represent 8% of a single soybean seed which comprise about 86% complex carbohydrates [130]. They are also comprised of 40 to 45% cellulose and 30 to 35% xylan on a dry basis [131]. High amount of xylan is believed to produce high level of xylanase activity. Gawande and Kamat [50] proved that the production of cellulase-free xylanase by *A. terreus* strain 5 and *A. niger* strain 44 using soybean hulls as substrate yielded 6.2 U/ml and 8.7 U/ml, respectively. Likewise, Kavya and Padmavathi [33] showed that soybean hulls possessed great potential as a substrate on the xylanase production using *A. niger* in SsF. In another study conducted by Brijwani and Vadlani [132], they observed that soybean hulls were a good substrate in the production of cellulolytic and xylanolytic enzymes including endoglucanase, β -glucosidase and xylanase. As a result, soybean hulls are suitable for fungal growth and enzymes production due to their high cellulosic composition [132].

Barley

Barley (*Hordeum vulgare*) is one of the major world crops today and it is grown worldwide due to its tremendous potential in industrial applications including food and animal feed, malting and brewing. The barley husk or 'cover' of barley is commonly removed during the pearling process. Barley grains comprise of two significant non-starch polysaccharides of 1,3-1,4- β -glucans and arabinoxylan that located within the endosperm and grain cell walls [133,134]. In this respect, barley husk contains approximately 30% hemicelluloses whereas xylan accounts for about 25.7g/100g of hemicelluloses in barley husk [135]. 1,3-1,4- β -glucanase hydrolyses 1,3-1,4- β -glucans while xylanase that secreted by the aleurone layer of barley acts in the depolymerisation of arabinoxylan within the cell wall during germination process. As a result, some transferable nutrient components were produced during degradation of cell wall. Hence, several studies have shown the usage of barley in the form of bran or straw for the fermentation of microorganisms to produce xylanase enzyme. In the latest study conducted by Soliman et al. [136], they remarked that barley husk as a substrate was able to show tremendous potential in xylanase production by both *A. niger* and *T. viride* which achieved the highest xylanase activity of 12.5 ± 0.13 U/g substrate and 11.0 ± 0.13 U/g substrate, respectively. In addition, an increased of xylanase activity by *A. niger* on barley husk from 12.5 ± 0.13 U/g substrate before optimisation to 42.0 ± 0.22 U/g substrate after optimisation [136]. Moreover, barley straw was being used by *Sclerotinia sclerotiorum* S2 to produce xylanase activity of 0.83 U/ml [137].

Sawdust

Sawdust is a waste product of wood processing which is constituted of 45 to 50% cellulose and 23 to 30% lignin. In fact, sawdust with high composition of beechwood is a good substrate for the production of xylanase enzyme [27]. Sawdust also plays a crucial role as carbon

source in xylanase production. Xylanase production was found to be optimum with 117.0 U/ml by *Arthrobacter spp.* MTCC 6915 [35] and 1.35 U/ml by *Penicillium chrysogenum* PCL501 [138], respectively. On the other hand, xylanase production was 0.65 and 1.26 U/ml by *A. niger* ANL 301 [139] and *Rhizopus oryzae* ATCC 9363 [140], respectively.

Maize

Maize or corn is a cereal crop that grown all over the world with the worldwide production about 785 million tons (IITA 2009). In a study

conducted by Goyal et al. [141], maize straw was the best carbon source for xylanase production if compared with jowar straw and barseem straw due to the fact that maize contained high level of hemicellulose. In addition, Doner and Hicks [142] also identified that purified maize bran contained high level of hemicelluloses, there was consisted of 67.5% of arabinoxylan, 22.5% of cellulose and 2.4% of protein. Table 5 shows the effect of different carbon sources of agricultural extracts on the xylanase production by different microorganisms.

Microorganism	Carbon source	Xylanase activity (U/mL)	Reference
<i>Arthrobacter spp</i>	Sawdust	117	Muragan et al. [35]
<i>Bacillus circulans</i>	Bagasse	8.4	Bocchini et al. [118]
<i>Bacillus pumilus</i>	Paddy husk	224.2	Kapilan et al. [143]
<i>Bacillus subtilis</i>	Wheat bran	410	Sanghi et al. [77]
<i>Bacillus spp.</i>	Oat spelt	52	Anuradha et al. [144]
<i>Aspergillus niger</i>	Maltose	17.8	Simoës et al. [145]
<i>Streptomyces chartreusis</i>	Corn cobs	334.34	Li et al. [146]
<i>Trichoderma viride</i>	Sorbitol	169	Simoës et al. [145]

Table 5: Effect of different carbon sources of agricultural extracts on the xylanase production by different microorganisms.

Culture conditions for xylanase production

Culture growth conditions including incubation temperature, pH and agitation speed play an important role in leading to greater xylanase production by various microorganisms.

Effect of incubation temperatures on xylanase production

The incubation temperature is one of the important parameters to determine the performance of xylanase fermentation. Generally, fungi including *Aspergillus spp* were cultivated at the temperature ranging from 25 to 45°C. Djekrif-Dakhmouche et al. [147] pointed out that *A. niger* ATCC 16404 was best cultured at 30°C for adequate growth to enhance the optimum liberation of xylanase. Likewise, in another study carried out by Ali et al. [148], they observed that the ideal growth temperature for *A. niger* GCBT7 was at 30°C and they also proved that 30°C was the optimum temperature for both enzyme activity and citric acid production in a stirred tank fermentor. Moreover, Maciel et al. [28] observed that 30°C was the optimum temperature for xylanase activity by *A. niger* LPB 326. These results were similar to the report by Shah and Madamwar [36] which stated that 30°C was the optimum temperature for xylanase production by *A. foetidus*. Likewise, *A. niger* B03 was best cultured at 29°C [149], *A. niger* LPB 326 was optimally cultured at 30°C ± 1 [28], *A. niger* KKS was at 30°C [150] and *A. niger* 44 was at 35°C [50], respectively. On the other hand, the ideal temperature for *A. niger* An-1.15 to acquire the optimum xylanase activity was observed to be at 28°C [151]. The optimum growth temperature for *A. niger* USM AII was at 28 ± 3°C which was similar to the optimum temperature for xylanase production [9]. Kavya and Padmavathi [33] showed that *A. niger* grown well at 28°C with the xylanase production of 8.98 U/ml. They also remarked that the cultivation temperature possessed huge impact to both xylanase production and growth of *A. niger*. Notably, *A. sulphureus* that isolated from soil in Northern China showed the optimum temperature for xylanase production occurred at the range

of 30 to 35°C [152], *A. foetidus* MTCC 4898 grown best at 30°C [36,153], *A. niger* FGSCA733 at 25°C [154], *A. nidulans* CECT 2544 at temperature ranging from 37 to 42°C [155], *A. carneus* M34 at 50°C [65], *A. niger* 44 at 35°C [50], *A. niger* isolated by preliminary screening at 28°C, respectively [33].

Effect of different initial pH medium on xylanase production

The pH medium has a huge influence on the performance of microbial xylanase activity where it plays a significant part in initiating the excretion of xylanase enzyme. *A. niger* is able to thrive in a slightly acidic environment of pH 6.5. In fact, majority of fungi are able to thrive at pH ranging from 5.0 to 8.0. Slightly acidic pH of 6.0 was observed by Maciel et al. [28] as the most suitable pH medium for fungi to flourish during fermentation process. Djekrif-Dakhmouche et al. [147] discovered that the best initial pH range for *A. niger* was noted to be at 5 or 6 while *A. niger* B03 was at pH ranging from 6.2 to 6.4 [149]. *A. fumigatus* AR1 was observed to produce the maximum xylanase activity at pH 6.0 to 6.5 [156], *A. nidulans* CECT 2544 at pH 6.8 [155] and *A. carneus* M34 at pH 6, respectively [65]. Additionally, low pH value inhibits the growth of other microorganisms particularly bacteria. Maciel et al. [28] observed that *A. niger* LPB 326 achieved optimal growth at low pH level around pH 6.0. Meanwhile, according to Springer [157], *A. ochraceus* grown well at pH level between pH 3 and 10, while slower growth was anticipated at pH 2.2. Some isolated *A. niger* is natural acidophilic fungi which grown well at pH 4.5 to 4.8 [158]. In addition, Jayant et al. [159] observed that *A. niger* in their study grown well at the slight acidic pH 5. Likewise, *Penicillium implicatum* and *Fusarium solani* produced the maximum xylanase activity of 58.80 and 28.60 U/mL at pH medium 5.5 and 6, respectively.

Effect of different agitation speeds on xylanase production

Agitation speed plays a very significant role in the fermentation process because it affects the dissolved oxygen and mass transfer of cultivation medium [47,160]. Filamentous fungi require appropriate agitation speed for uniform distribution of the spores and prevention of the emergence of inactive clump of fungal growth. In a study established by Palma et al. [161], a low xylanase activity was obtained due to shear stress occurred at higher agitation speed of 400 per minute during the growth of fungi *Penicillium janthinellum* that isolated from decaying wood. On the contrary, the most desirable agitation speed to produce high xylanase enzyme was achieved with a speed of 60 per minute in some studies. Another study carried out by Purwanto et al. [160], the optimal growth of *A. niger* was occurred at agitation speed of 100 rpm. However, when the agitation speed increases to 200 rpm, it causes higher shear stress and mechanical forces disruption on filamentous fungi and thus results in the formation of small pellet size of *Aspergillus spp.* Meanwhile, a study by Fang et al. [65] demonstrated that the optimum xylanase production of 22.2 U/mL by *A. carneus* M34 was observed at agitation speed of 111.9 rpm when utilising oat spelt xylan as the carbon source. On the contrary, Haltrich et al. [13,31] stated that the static growth condition was more suitable and preferable for fungi. His statement was supported by Djekrif-Dakhmouche et al. [147] that during fermentation process of *A. niger* ATCC 16404, there was no significant results detected on the production of the desirable enzyme at agitation speed of 150 to 200 rpm. Nevertheless, Darah et al. [162] remarked that the optimum agitation speed for the maximum growth of *A. niger* FETL FT3 of 3.75 g/L was observed at 200 rpm. There are number of different agitation speeds used by different researchers to obtain the maximum growth of *Aspergillus spp.* in fermentation process such as 100 rpm [163,164], 130 rpm [37], 180 rpm [9] and 200 rpm to achieve their optimal xylanase production [148,162].

Conclusion

The development of enzyme industries are emerging and growing rapidly in the recent decade. In fact, the market of xylanase is expanding rapidly due to its greater potential in industrial uses, particularly in the biotechnological applications. Xylanase possesses growing demand globally because of its vast applications in wide industries ranging from pulp and paper to textile industry. Xylanase produced from different fungi are found to be more satisfactory in terms of its activity which is easily determined by DNS method. Agricultural extracts or lignocellulosic materials are commonly utilised as carbon source in medium formulation because they are plenteous in nature, low in cost with high level of carbohydrate content which are suitable to encourage production of xylanase by various microorganisms especially *Aspergillus* and *Bacillus spp.* Moreover, agricultural extracts are the important raw materials resource and energy source to enhance the growth of microorganisms. They are basically constituent of cellulose, hemicellulose and lignin. Indeed, they contain high concentration of xylan which consisted of hemicellulose is potent as an inducer for xylanase production. In addition, utilisation of agricultural extracts such as wheat bran, rice bran, palm kernel cake, sugarcane baggase, maize, barley husk, soybean hulls and sawdust is notably economical and environmentally sound. Besides that, culture growth conditions including incubation temperature, pH medium and agitation speed also play a crucial role in the optimisation of growth of microorganisms and xylanase production. Nonetheless, different microorganism strains isolated

from different origins around the world might have slightly differences in their growth conditions for xylanase production. All in all, xylanase possesses better potential in the coming future as one of the valuable enzymes in various applications specifically in pulp and paper industry as well as bioethanol production.

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