Hemicellulose is part of the core constituents of lignocellulose and it is the next most abundant carbohydrate resource on Earth after cellulose (Radhika et al. 2011). Xylan represents the main component of hemicelluloses (Bajaj and Manhas 2012) and is the major backbone in the hemicellulosic portion of the plant cell wall, connecting compounds like arabinose, mannose, glucose and other sugars via an acetyl chain (Radhika et al. 2011). Xylan is a heterogeneous or diverse polysaccharide and contains a linear backbone of β-1,4-D-xylopyranoside residues and, also short side-chain branches. Xylan can only be completely hydrolyzed using several enzymes due to its composite structure (Bajaj and Manhas 2012). Thus, biodegradation of xylan, requires series of xylanolytic enzymes among which endo-β-1,4-xylanases (EC 3.2.1.8) hydrolyse the xylan backbones into short xylooligosaccharides (Pandey et al. 2014).

Extracellular xylanases synthesized by microorganisms have incredible industrial significance, with about 20% of global enzyme market being shared by cellulase, pectinase, and xylanase (Polizeli et al. 2005). The global market of industrial enzymes is said to have witnessed rapid growth in recent years. It was reported to be only 1 million US dollars in 1970 and has grown to 4.5 billion dollars in 2012, with projection to reach about 7.1 billion dollars in 2018 (Kalim et al. 2015).

Microbial enzymes, like xylanases are imperative in numerous fields from food processing to paper and pulp industries (Azin et al. 2007). It contributes a critical role in numerous biotechnological processes such as clarification of fruit juice, bleaching, beer and wine, improving poultry feed digestibility, pulp and paper, leather and baking industries (Uday et al. 2016a). They are utilized in extraction of extracellular polymeric substances (EPS) and plant oils and, also used to enhance the nutritional quality of silage, coffee, green feed and starch (Lakshmi et al. 2009). Moreover, an important usefulness of xylanase has been established to produce biofuel from lignocelluloses (Uday et al. 2016b). Its use most recently reported for the green synthesis of nanoparticles with profound biomedical applications (Elegbede et al. 2018a, 2018b, 2019) has further expanded the frontiers of enzyme technology in nanobiotechnology (Lateef and Adeeyo 2015; Lateef et al. 2015; Adelere and Lateef 2016). Broad types of fungi and bacteria are known to synthesize xylanases under various cultivation systems, but filamentous fungi are the most effective producers of xylanase, because they

**Optimization of the production of xylanases in corncob-based media by Aspergillus niger and Trichoderma longibrachiatum using Taguchi approach**

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**ABSTRACT** Xylanases are important in producing several commercially valued bio-products. In this study, xylanases were produced by *Aspergillus niger* L3 and *Trichoderma longibrachiatum* L2 using corncob, an agricultural waste, as sole carbon source. The impact of important fermentation parameters at individual and interactive levels were studied using Taguchi L9 orthogonal array. Substantial variation in enzyme synthesis was observed among designated factor levels. The optimal conditions to produce xylanases were 20% inoculum size, 24 h fermentation time, substrate concentration of 15 g/l at pH 5.5 for *A. niger* L3; and inoculum size 12.5%, 72 h fermentation time, substrate concentration of 15 g/l at pH 5.5 for *T. longibrachiatum* L2. Validation of outcomes of the optimal combination of parameters resulted in a significant improvement of approximately 208.09 and 192.59% in the yield of xylanase by *A. niger* L3 (28.69 to 88.39 U/ml) and *T. longibrachiatum* L2 (22.13 to 64.75 U/ml), respectively. The study therefore established the optimal valorization of corncob to produce xylanase by the fungal isolates.

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**KEY WORDS** agrowastes, corncob valorization, optimization, Taguchi orthogonal array, xylanase

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secretes elevated levels of enzymes compared to yeasts and bacteria (Rani et al. 2014). On an industrial scale, xylanases are principally synthesized by *Aspergillus* and *Trichoderma* spp. (Shahi et al. 2011). Also, Aspergilli have capability to synthesize broad range of enzymes responsible for degrading plant cell wall. *Trichoderma* spp., which are foremost agents causing decay and decomposition of agricultural wastes possess an array of different enzymes, and hence they are known as excellent producers of lignocellulolytic enzymes (Azin et al. 2007).

Pure substrates are highly expensive and are not affordable for bulk production of enzymes at industrial level. Therefore, it is essential to survey cheap substrates for cost-effective production of enzymes. Agricultural residues, including animal wastes present a cheap source of raw materials for the industrial production of enzymes (Geetha and Gunasekaran 2010; Lateef et al. 2008, 2010, 2012; Lateef and Gueguim-Kana 2012; Ganaie et al. 2014; Lateef et al. 2015), and they are available in great quantity in countries with wide-ranging agricultural practices (Bajaj and Manhas 2012).

Optimization of production of xylanase is a prerequisite for its large-scale economical production (Azin et al. 2007), and this can be achieved by manipulating important parameters that affect the fermentation process (Rani et al. 2014). Production of xylanase differs in diverse strains and this can be regulated by the nutritional, physiological, and biochemical nature of microbes that are employed (Lakshmi et al. 2009). In this case, it becomes imperative to optimize all fermentation parameters since no definite medium has been authenticated for the maximum production of any metabolite, because the genetic diversity in various microbial sources causes each microorganism or strain to have its unique conditions for the best yield of production (Rao et al. 2008a). Notable fermentation and environmental factors that can manipulate metabolism-mediated yields include temperature, aeration, pH, agitation, carbon and nitrogen sources, incubation time, initial inoculum size, ion requirement amongst others (Prakasham et al. 2007a). Hence, for industrial and commercial production, optimization of growth medium is one of the critical steps to reduce the quantity of unutilized constituents for a cost-effective yield (Lakshmi et al. 2009).

The conventional optimization procedures, including one-factor-at-a-time design entail experimental work which is time consuming and does not provide necessary information about the common interactions of the parameters (Mandal et al. 2015). On the contrary, statistical optimization techniques help to investigate the influence of controlled factor in a multivariate system. Furthermore, bioprocesses have been optimized using response surface methodology (RSM), artificial neural networks (ANN), and genetic algorithm (Gueguim-Kana et al. 2007, 2012a,b; Adeoye et al. 2015; Adeyo et al. 2016).

Taguchi orthogonal array (OA) design of experiment (DOE) encompasses the investigation of a system by a set of independent variables (factors or parameters) over specific levels of interest (Taguchi 1986). The technique also determines the relationship or correlation between variables and operational conditions (Mandal et al. 2015). Recent literature reviews reveal that Taguchi methodology has been used to optimize reaction variables in several biochemical processes by studying a given set of independent variables, which may be controllable or uncontrollable over a definite region of interest. It is also imperative that experiments conducted in small scale are valid over an entire experimental region (Uday et al. 2016a). The use of ANOVA (analysis of variance) is to investigate the accuracy of experimental data and gives the statistical relationship of the output (Mandal et al. 2015). Taguchi methodology has been efficiently applied in bioprocesses (Rao et al. 2008b), to optimize the synthesis of some industrial enzymes such as tannase (Mohapatra et al. 2009), alkaline protease (Laxman et al. 2005), α-amylase (Uysal et al. 2010) and L-asparaginase (Prakasham et al. 2007b). It is better and superior over other analogous statistical designs, including the RSM, because much less time is needed to execute the experiment. Also, Taguchi method is advantageous to achieve consistency and reliability at little cost with fewer experiments, when compared with RSM and ANN. Its application normally leads to improved quality or yield of products and process performance (Rao et al. 2008b). The improved performance is ensured by the orthogonal layout, whereby interactive effects of different factors are studied with fewer numbers of experimental runs (Rao et al. 2008b). However, Taguchi DOE does not take into accounts of the influence of all the controlling factors in a process, but chiefly concerned about the main effects of the important factors. Also, its application requires a critical brainstorming on the discovery of key parameters, and competence in statistical analysis. It is also expedient that for reliability, the noise factors in the experiment must be properly identified (Rao et al. 2008b).

Most recently, we reported xylanase activities of some fungal isolates, including *Aspergillus niger*, *A. flavus*, *Trichoderma longibrachiatum*, *A. fumigatus*, *Fusarium solani*, and *Botryodiplodia* sp. in our laboratory with potent dough rising and juice clarification activities (Elegbede and Lateef 2018). In the present study, Taguchi method was applied to optimize process parameters to synthesize xylanase by two of the isolates; namely *A. niger* L3 and *T. longibrachiatum* L2 in corncob-based media with the aim of improving the enzyme yield for biotechnological applications. The experiments were designed using 4 factors at 3 levels with OA layout of L9 (3⁴).
Materials and Methods

Microorganisms and maintenance
Isolates of A. niger L3 and T. longibrachiatum L2 which were previously isolated in our laboratory (Elegbede and Lateef 2018) were used in this study. They were sub-cultured on fresh sterile potato dextrose agar (PDA) plates, incubated at room temperature (30 ± 2 °C) for 72 h and stored on PDA at 4 °C.

Substrate
Corncobs were locally sourced from Ajegunle market, Oyo, Oyo state, Nigeria, and processed through grinding (0.5 mm mesh) to obtain fine powder. The percentage moisture content of the corncob powder was estimated by drying the powder to a constant weight at 110 °C in a hot air oven (Lateef and Gueguim-Kana 2012).

Inoculum preparation
Inoculum was developed according to the methods of Lateef and Gueguim-Kana (2012) through the transfer of a loopful mycelium of the fungal strain from the plate into a sterilized 50 ml inoculum medium in a 250 ml capacity flask (containing 1% sucrose, 0.2% yeast extract, final pH 5.5). The flask was incubated at 30 ± 2 °C on a shaker at 100 rpm for 24 h.

Submerged fermentation
The basal medium composition for enzyme production was corncob (20.0 g/L), MgSO₄ (2.0 g/L), NaNO₃ (1.4 g/L), KH₂PO₄ (1.8 g/L), NH₄Cl (2.0 g/L), and CaCO₃ (1.2 g/L) at starting pH of 5.7 as previously reported (Elegbede and Lateef 2018). Then, 40 ml of basal medium was dispensed in 250 ml flask, sterilized at 121 °C for 15 min and inoculated with 10% (v/v) 24 h old inoculum (5 × 10⁶ spores per ml). The culture was incubated at 30 ± 2 °C on a shaker at 100 rpm for up to 120 h. After specific period of fermentation, the contents were filtered with the aid of Whatman No.1 filter paper, followed by centrifugation at 4000 rpm at 10 °C for 25 min. The cell-free supernatant was used as crude enzyme and kept at 4 °C until further use.

Xylanase assay
Xylanase activity was determined following the methods of Bailey et al. (1992). Exactly 0.9 ml of 0.5% (w/v) of beechwood xylan (Megazyme, Ireland) prepared in 0.1 M sodium citrate buffer at pH 5.4 was reacted with 0.1 ml of the crude enzyme, and incubated in a water bath shaker at 50 °C for 30 min. The reaction was terminated by adding 1 ml of 3,5-dinitrosalicylic acid reagent (DNS acid) to the reaction mixture followed by heating at 100 °C for 10 min. This was followed by cooling to room temperature and subsequent measurement of the absorbance at 540 nm using Spectrumlab S23A spectrophotometer (Yuchengtech, Beijing City, China). Earlier, xylose standard curve was prepared for the extrapolation of the amount of xylose released through the enzymatic hydrolysis of xylan. A unit (U) of xylanase was defined as the amount of enzyme that liberated 1.0 μmol of reducing sugar as xylose equivalent per minute in the reaction mixture as described in the above assay conditions. All enzyme determinations were investigated in duplicates.

Taguchi optimization
Taguchi method was employed in investigating the relationships that exist among the variables of medium components and to optimize for higher yields of enzyme production. Taguchi design methodology was performed using modified steps as stated in Figure 1, and it was used to set up the critical fermentation factors: inoculum size (%), fermentation time (h), initial pH, and substrate (corncob) concentration (g/L). The effective levels were set as low, intermediate and high (Table 1) based on earlier experimental values (Elegbede and Lateef 2018). Conditions such as temperature (30 ± 2 °C), agitation (100 rpm), and media: flask volume ratio (40 ml/250 ml flask) were not changed and fixed as standard conditions for the fermentation.

Table 1. Culture parameters and their respective levels of variation.

| Culture parameters | Units | Levels of variation |
|--------------------|-------|---------------------|
| Inoculum size      | %     | 5 12.5 20           |
| Fermentation time  | h     | 24 72 120           |
| Substrate (corncob)| g/l   | 5 15 25             |
| Initial pH         |       | 4 5.5 7             |

The appropriate Taguchi design arrangement for data analysis was selected. The L9-orthogonal array method
based on modified methods described by Taguchi (1986), which overcomes many problems related with conventional methodology, was used for the aforementioned control parameters with three levels of factor variation. In Taguchi method, the summary statistic $\eta$ is called signal-to-noise ratio (SNR). The value of $\eta$ is an effective indicator for the assessment of the impact of process parameters on enzyme synthesis. The larger-the-better signal-to-noise was utilized to evaluate the summary statistic $\eta$ in the present study, because an improved production of xylanase was targeted, and it was defined as follows:

$$SNR (\eta) = -10 \log (R^{-2})$$

Eqn. 1. (Chou et al. 2003)

Where, SNR represents the signal-to-noise ratio, while R represents the response or yield of correspondent trials. For each component, the optimal conditions give the largest SNR ratio.

The optimal combination of fermentation parameters was selected using the effect table, and the result was predicted using the formula (Eqn. 2). To validate or confirm the optimized methodology, fermentation experiments were run in duplicates and the samples collected were assayed for xylanase production. ANOVA was performed to evaluate the fermentation parameters that were statistically significant.

$$y_{pr} = y_{av} + \sum_{i=1}^{n-1} (y_{opt} (n) - y_{ave})$$

Eqn. 2

**Results and Discussion**

**Unoptimized xylanase production**

Xylanase was produced by *A. niger* L3 and *T. longibrachiatum* L2 strains on corncob supplemented minimal salt media since the use of pure xylan for industrial production of xylanases is uneconomical because of its high cost. Hence, use of cost-effective xylan-rich substrates is highly recommended (Lakshmi et al. 2009). The two fungi utilized corncob effectively as potential source of carbon to produce xylanase (Elegbede and Lateef 2018). Carbon source has been well-known as a noteworthy factor during the growth and metabolic process of any

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**Table 2.** Experimental set-up (L9 orthogonal array) for xylanase production by *A. niger* L3 and *T. longibrachiatum* L2.

| Exp. no | Inoculum size | Fermentation time | Substrate concentration | Initial pH | Xylanase activity (U/ml) | Signal-to-noise ratio | Xylanase activity (U/ml) | Signal-to-noise ratio |
|---------|---------------|-------------------|-------------------------|------------|--------------------------|-----------------------|--------------------------|-----------------------|
|         |               |                   |                         |            | A. niger                 |                       | T. longibrachiatum       |                       |
| 1       | 1             | 1                 | 1                       | 1          | 18.44                    | 25.31522              | 16.53                    | 24.36546              |
| 2       | 1             | 2                 | 2                       | 2          | 53.28                    | 34.53128              | 56.83                    | 35.09155              |
| 3       | 1             | 3                 | 3                       | 3          | 12.57                    | 21.98671              | 4.51                     | 13.08353              |
| 4       | 2             | 1                 | 2                       | 3          | 39.07                    | 31.83687              | 55.87                    | 34.94357              |
| 5       | 2             | 2                 | 3                       | 1          | 1.50                     | 3.521825              | 7.24                     | 17.19477              |
| 6       | 2             | 3                 | 1                       | 2          | 41.53                    | 32.36724              | 43.45                    | 32.75980              |
| 7       | 3             | 1                 | 3                       | 2          | 62.43                    | 35.90787              | 6.15                     | 15.77750              |
| 8       | 3             | 2                 | 1                       | 3          | 31.69                    | 30.01844              | 59.16                    | 35.44056              |
| 9       | 3             | 3                 | 2                       | 1          | 43.58                    | 32.78574              | 47.68                    | 33.56672              |

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**Table 3.** Effect table of xylanase production by *A. niger* L3 and *T. longibrachiatum* L2.

| Levels of parameters | A. niger L3 | Parameters | T. longibrachiatum L2 |
|----------------------|-------------|------------|------------------------|
| Inoculum size        | Fermentation time | Substrate concentration | Initial pH |
| 1                    | 27.27774    | 31.01998   | 29.23363               | 20.54093   |
| 2                    | 22.57531    | 22.69052   | 33.05130               | 34.26880   |
| 3                    | 32.90402    | 29.04656   | 20.47213               | 27.94734   |

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**Table 4.** Experimental set-up (L9 orthogonal array) for xylanase production by *A. niger* L3 and *T. longibrachiatum* L2.

| Exp. no | Inoculum size | Fermentation time | Substrate concentration | Initial pH | Xylanase activity (U/ml) | Signal-to-noise ratio | Xylanase activity (U/ml) | Signal-to-noise ratio |
|---------|---------------|-------------------|-------------------------|------------|--------------------------|-----------------------|--------------------------|-----------------------|
|         |               |                   |                         |            | A. niger                 |                       | T. longibrachiatum       |                       |
| 1       | 1             | 1                 | 1                       | 1          | 18.44                    | 25.31522              | 16.53                    | 24.36546              |
| 2       | 1             | 2                 | 2                       | 2          | 53.28                    | 34.53128              | 56.83                    | 35.09155              |
| 3       | 1             | 3                 | 3                       | 3          | 12.57                    | 21.98671              | 4.51                     | 13.08353              |
| 4       | 2             | 1                 | 2                       | 3          | 39.07                    | 31.83687              | 55.87                    | 34.94357              |
| 5       | 2             | 2                 | 3                       | 1          | 1.50                     | 3.521825              | 7.24                     | 17.19477              |
| 6       | 2             | 3                 | 1                       | 2          | 41.53                    | 32.36724              | 43.45                    | 32.75980              |
| 7       | 3             | 1                 | 3                       | 2          | 62.43                    | 35.90787              | 6.15                     | 15.77750              |
| 8       | 3             | 2                 | 1                       | 3          | 31.69                    | 30.01844              | 59.16                    | 35.44056              |
| 9       | 3             | 3                 | 2                       | 1          | 43.58                    | 32.78574              | 47.68                    | 33.56672              |

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**Table 5.** Effect table of xylanase production by *A. niger* L3 and *T. longibrachiatum* L2.

| Levels of parameters | A. niger L3 | Parameters | T. longibrachiatum L2 |
|----------------------|-------------|------------|------------------------|
| Inoculum size        | Fermentation time | Substrate concentration | Initial pH |
| 1                    | 27.27774    | 31.01998   | 29.23363               | 20.54093   |
| 2                    | 22.57531    | 22.69052   | 33.05130               | 34.26880   |
| 3                    | 32.90402    | 29.04656   | 20.47213               | 27.94734   |

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Optimization of the production of xylanases

Table 4. Performance of selected optimal combination of parameters.

| Parameters                  | Levels         |
|-----------------------------|----------------|
|                             | A. niger L3    | T. longibrachiatum L2 |
| Inoculum size (%)           | 20             | 12.5                  |
| Fermentation time (h)       | 24             | 72                    |
| Substrate concentration (g/L)| 15             | 15                    |
| Initial pH                  | 5.5            | 5.5                   |
| Optimum profile             | A3B1C2D2       | A2B2C2D2              |
| *Signal-to-noise ratio      | 38.92731       | 36.220975             |
| *Xylanase activities (U/ml) | 88.39          | 64.75                 |

*Average of two readings. A: inoculum size, B: fermentation time, C: substrate concentration, D: initial pH

microorganism; therefore, the choice of a suitable carbon source has been identified as a key determinant in the production economics of xylanase (Pandya and Gupte 2012). Xylanase production was performed in 250 ml Erlenmeyer flasks containing 40 ml of fermentation medium as earlier stated. The time course analysis of xylanase activities showed the commencement of production of xylanase at 24 h of fermentation for both isolates and reached maximum of 28.69 U/ml at 96 h for A. niger L3, and 22.13 U/ml at 72 h for T. longibrachiatum L2 as previously reported (Elegbede and Lateef 2018). These productivities were achieved at pH values of 6.7 and 6.6 for A. niger and T. longibrachiatum L2, respectively. Maximum yields of fungal xylanases have been achieved at pH of 4.0-6.5 in similar studies (Murthy and Naidu 2012; Yegin 2017).

Optimization of xylanase production through Taguchi method

Enhancement of production of metabolites by microbes is influenced by the nutritional, physiological, and biochemical nature of the microbe utilized, and these influential factors vary from microorganism to microorganism (Lakshmi et al. 2009). The Taguchi L9 orthogonal array revealed noteworthy variation in xylanase synthesis (Table 2). Initial maximum xylanase production values were observed to be 62.43 and 59.16 U/ml for A. niger L3 and T. longibrachiatum L2, respectively. These results are regarded as the local optimal for xylanase production by both fungal strains. The data presented a very significant improvement in xylanase yields compared to the conventional unoptimized production; 117.60 and 167.33% for A. niger L3 and T. longibrachiatum L2, respectively. Similar variation of enzyme activity was reported by Lakshmi et al (2009). This data further showed that xylanase production by fungal strains is determined by fermentation parameters and their levels.

The Taguchi DOE used for optimization in this study provided the predicted profile for maximum yield of xylanase from the effect tables. The S/N ratio was used to determine the influence of each variable on the output, and the effect table (Table 3) was employed for the analysis of the relative effect of different parameters. This is because the change in signal leads to a larger effect on the output variable being evaluated. The S/N ratio values help in the assessment of the combination of factors that have the maximum influence on the response characteristic of concern, such that higher values of S/N ratio indicate greater influence of the combined parameters (Rani et al. 2012). The profiles A3B1C2D2 (interpreted as inoculum; 20%, fermentation time; 24 h, substrate concentration; 15 g/L, and pH; 5.5) and A2B2C2D2 (interpreted as inoculum; 12.5%, fermentation time; 72 h, substrate concentration; 15 g/L, and pH; 5.5) were generated for A. niger L3 and T. longibrachiatum L2, respectively, being the optimum conditions for achieving higher xylanase yields (Table 4).

The optimal combination of parameters was evaluated and the verification experiments, in which optimized conditions were used yielded highest activities of 88.39 and 64.75 U/ml for A. niger L3 and T. longibrachiatum L2, respectively as the global maximum (Table 4). These global maxima obtained from the optimum combination profiles represented 41.58 and 9.45% increase on the local maxima obtained from the initial orthogonal array experiments. The results indicated a total significant increment of 208.09 and 192.59% when compared with the conventional unoptimized xylanase synthesis by both A. niger L3 and T. longibrachiatum L2, respectively. These results compared well with previous reports of xylanase production that were optimized using Taguchi method where 277%, 161.5%, 41.9 and 10.24% higher enzyme yields were obtained (Lakshmi et al. 2009; Rani et al. 2014; Mandal et al. 2015). The increase can be claimed to have resulted from the interactions and relations among the various factors (Mandal et al. 2015).

Analysis of Variance (ANOVA)

Understanding of the influence or effect of each individual factor is crucial for the implementation of a successful bioprocess operation (Rani et al. 2014). Therefore, ANOVA was employed in evaluating results of the orthogonal array experiment and to study the level of consequence of the fermentation parameters on the deviation of the responses (Lakshmi et al. 2009). ANOVA (Table 5) showed that the initial pH, substrate concentration, inoculum size and fermentation time contributed 35.10, 30.93, 19.88 and 14.09%, respectively, to the optimal yield of xylanase by A. niger L3, while substrate concentration, inoculum size, fermentation time and initial pH contributed 88.99, 4.81, 3.94 and 2.26%, respectively, to the optimal yield of
xylanase in the case of *T. longibrachiatum* L2. While initial pH has the highest effect on yield of xylanase by *A. niger* L3, it has the least effect and a very minute significance on *T. longibrachiatum* L2. This further shows that factors that influence the production of metabolites vary from microorganism to microorganism which can be adduced to genetic diversity, physiology and metabolic differences. The inference of this finding is that more interest should be focused on initial pH and substrate concentration for *A. niger* L3, and mainly substrate concentration for *T. longibrachiatum* L2, if the production of xylanases by any of the fungal isolates is to be significantly adjusted or altered. It can therefore be concluded that corncob can be valorized to produce xylanase by the fungal isolates.

**Conclusion**

Xylanase production by *A. niger* L3 and *T. longibrachiatum* L2 on corncob based media was successfully enhanced through optimization of the fermentation conditions with Taguchi method. Four factors (Inoculum concentration (%), substrate concentration (g/l), fermentation time and initial pH) at three levels of variation were used and the analysis confirmed the participation and, also the interactions of the factors. It was ascertained that the optimal conditions for production of xylanases by *A. niger* L3 and *T. longibrachiatum* L2 were inoculum; 20%, fermentation time; 24 h, substrate concentration; 15 g/l, and pH; 5.5 and inoculum; 12.5%, fermentation time; 72 h, substrate concentration; 15 g/l, and pH; 5.5, respectively. Using the optimum conditions, enzyme yield was significantly improved to about 208.09 and 192.59% for *A. niger* L3 and *T. longibrachiatum* L2, respectively. Also, it was established that more attention should be focused on initial pH and substrate concentration for *A. niger* L3, and mainly substrate concentration for *T. longibrachiatum* L2, if the production of xylanases by any of the fungal isolates is to be significantly adjusted or altered. It can therefore be concluded that corncob can be valorized to produce xylanase by the fungal isolates.

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