



Detergent-compatible fungal cellulases

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Received: 12 June 2020 / Accepted: 5 November 2020 / Published online: 12 November 2020
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Abstract

Detergent enzymes are currently added to all powder and liquid detergents that are manufactured. Cellulases, lipases, amylases, and proteases are used in the detergency to replace toxic phosphates and silicates and to reduce high energy consumption. This makes the use of enzymes in detergent formulation cost effective. Fungi are producers of important extracellular enzymes for industrial use. The fungal and bacterial cellulases maintain the shape and color of the washed garments. There is a high demand for cellulases at the market by detergent industries. With this high demand, genetic engineering has been a solution due to its high production of detergent-compatible cellulases. Fungi are the famous source for detergent-compatible cellulases production, but still, there is a lack of the cost-effective process of alkaline fungal cellulase production. Review papers on detergent-compatible bacterial cellulase and amylase and detergent-compatible fungal and bacterial proteases and lipases are available, but there is no review on detergent fungal cellulases. This review aims to highlight the production, properties, stability, and compatibility of fungal cellulases. It will help other academic and industrial researchers to study, produce, and commercialize the fungal cellulases with good aspects.

Introduction

Cellulase is an hydrolase enzyme, cleaving β -1,4-glycosidic bonds of cellulose or its derivatives like cellooligosaccharide, to glucose monomers. To completely hydrolyze these polymers to glucose units, three enzymes act synergistically. These are endoglucanases (EC 3.2.1.4) that break down internal glycosidic bonds, exoglucanases (EC 3.2.1.91) cleaving chain termini liberating cellodextrins, and β -glucosidases (EC 3.2.1.21) that liberate glucose units following cellodextrins degradation (Uhlig 1998; Sajith et al. 2016). Fungi are the microorganisms of choice to produce industrial cellulases owing to desired properties like extracellular secretion of an enzyme in huge amounts with cost-effective substrates (Bhat 2000; Ahmed and Bibi 2018). Detergent-compatible cellulases are easily obtained abundantly from fungi compared to plants and animals (Acharya and Chaudhary 2012). In addition, the genetic material of fungal species is easily cloned into the bacterial strain for overproduction of cellulases because fungal cellulases are less complex in structure compared to

bacterial ones (Uhlig 1998; Maki et al. 2009; Acharya and Chaudhary 2012).

Cellulases are used in different industries such as detergent, biofuel, food, textile, cosmetics, feed, chemicals, pulp, and paper (Acharya and Chaudhary 2012; Juturu and Wu 2014; Sajith et al. 2016). The fungal cellulases are importantly used in the detergent preparation to aid in the defibrillation processes. They increase the color brightness/softness of the fabric and minimize the defibrillation cost (Miettinen-Oinonen and Suominen 2002; Koga et al. 2008). Oxidizing agents, anionic and non-ionic detergents, are the constituents of a detergent. The bacterial and fungal enzymes are stable and resistant to oxidizing agents (Niyonzima and More 2014c). In the detergency, the stability at alkaline pH is also a prerequisite for fungal cellulases (Miettinen-Oinonen and Suominen 2002; Koga et al. 2008; Niyonzima 2019). For instance, the cellulase of *Humicola insolens* that was active in the alkaline region of pH 8.5–9.0 and at temperature of 50 °C was used in washing powder to remove soil-related dirtiness. It was then commercialized due to these better properties (Uhlig 1998; Behera et al. 2017). Therefore, during washing, cellulases hydrolyze effectively sebum stain between inter-microfiber spaces without any fabric damage. Indeed, the inter-microfibers arise from mechanical damage during washing (Boisset et al. 1997; Uhlig 1998).

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Enzymes used in the detergent industry may be immobilized. In the immobilization process, free cells or enzymes like cellulase are confined to an inorganic/organic or hybrid carrier to improve some properties. For instance, an improved washing capacity of commercial cellulase to remove indigo dye from denim fabrics was observed, compared to free cellulases, after enzyme immobilization on ZrOCl₂-activated pumice support (Pazarlioglu et al. 2005). Zdartar et al. (2017) synthesized a lignin-TiO₂ hybrid carrier. An increase of cellulase activity was observed for the cellulase purified from *Aspergillus niger* when immobilized to this support. Similarly, compared to free cellulase, the higher activity and stability of *A. niger* cellulase were observed when the enzyme was immobilized on a sponge through the glutaraldehyde as spacer group (Ahmed et al. 2013). Although cellulases immobilization can play a role in improving washing performance, the process should be checked for the cost-effectiveness.

Cellulases are utilized in detergent preparation along with other enzymatic detergents including fungal or bacterial proteases, amylases and lipases. The combination of these four enzymatic detergents facilitates the removal of all soil-related dirtiness and maintains the quality of all fabrics. For example, the SaniZyme® contains all these 4 detergent enzymes and removes various stains such as proteins, blood, carbohydrates, lipids, and mucous from the medical instrument because it is manufactured as a liquid detergent. Getinge Clean MIS Detergent® is another complex detergent enzyme; when combined with other components like surfactants at pH 8.0, it cleans all types of soils (Boisset et al. 1997; Uhlig 1998; Jayasekara and Ratnayake 2019).

Although branded cellulases such as Carezyme®, Celluzyme®, and SaniZyme® are available at the market for washing of fabrics and garments, they do not meet all the requirements. Therefore, the search for fungal cellulases with desirable aspects has to continue. The review papers on the detergent-compatible bacterial amylases and cellulases and detergent-compatible microbial proteases and lipases are available (Niyonzima and More 2014a; Niyonzima and More 2015a, b, c; Niyonzima 2018), but there is a gap for a dedicated report for detergent fungal cellulases. In this article, the production, properties, and compatibility of detergent fungal cellulases are addressed to the worldwide researchers.

Isolation and identification of detergent-compatible cellulases produced by fungi

Most of the detergent-compatible cellulases are screened from soil (Dave et al. 2015; Maharana and Ray 2015; Bagewadi et al. 2016; Bairagi 2016; Imran et al. 2018; El-Baroty et al. 2019). The other detergent-compatible enzymes such as bacterial and fungal proteases (Niyonzima and More 2015a), bacterial and fungal lipases (Niyonzima and More 2015a),

bacterial amylases (Niyonzima and More 2014a), and bacterial cellulases (Niyonzima 2018) were also mostly isolated from soil microorganisms. Indeed, it is well known that soil harbors billions of microorganisms. The *Aspergillus terreus* that produces detergent-compatible cellulase was isolated from effluent. Iqbal et al. (2011), Pham et al. (2012), and Trinh et al. (2013) also isolated fungi that were able to produce detergent-compatible cellulases (Table 1).

Carboxymethylcellulose (CMC) agar is one of the screening media for fungi that produce detergent-compatible cellulases (Pham et al. 2012; Trinh et al. 2013; Bairagi 2016; Imran et al. 2018) (Table 1). Lignocellulosic substrate agar was the best screening medium for detergent-compatible cellulase secretion by *T. viride* (Iqbal et al. 2011). Sometimes, fungal growth media such as potato dextrose agar (PDA) and Sabouraud dextrose agar (SDA) are supplemented to CMC (Dave et al. 2015; Bagewadi et al. 2016) or to the filter paper strip (El-Baroty et al. 2019), respectively, for the secretion of detergent-compatible cellulases. A stain such as Congo red or chloramphenicol is added to the culture medium for better visualization of the cellulase hydrolysis zone. For instance, Congo red was used to observe the hydrolysis zone for the cellulase of *Aspergillus terreus* strain AKM-F3 (Maharana and Ray 2015). Sometimes, chloramphenicol is supplemented to CMC agar to prevent the bacterial growth. For instance, Bekele et al. (2015) inhibited bacterial growth by adding chloramphenicol to the cultivation medium when producing detergent-compatible cellulase from *Aspergillus terreus*.

The fungi producing detergent-compatible cellulases are identified based on morphological characteristics and microscopic features (Iqbal et al. 2011; Bekele et al. 2015; Bairagi 2016; Imran et al. 2018). However, most of them are currently identified with the help of ITS rDNA/RNA region sequencing (Trinh et al. 2013; Dave et al. 2015; Maharana and Ray 2015; Bagewadi et al. 2016; El-Baroty et al. 2019) (Table 1). Similarly, bacteria secreting detergent-compatible cellulases were generally identified based on morphological and microscopic characteristics followed by genetic material sequencing (Niyonzima 2018).

Production of detergent-compatible cellulases by fungi

Fungi secrete extracellular enzymes like cellulases that are important for industrial use (Nazir et al. 2010; Acharya and Chaudhary 2012). They are preferred because they secrete large quantities of enzymes when fermented on cheap materials like agro wastes or byproducts (Ahmed and Bibi 2018). These fungal cellulases are mainly obtained from 2 genera, viz., *Aspergillus* (Nguyen and Quyen 2010; Pham et al. 2012; El-Hadi et al. 2014; Bekele et al. 2015; Maharana and Ray 2015; Imran et al. 2018; El-Baroty et al. 2019) and *Trichoderma* (Iqbal et al. 2011; Ahmed et al. 2016;

Table 1 Fungal species identified as detergent cellulase producers

Fungal species	Isolation place	Screening solid medium	Identification methods	Reference
<i>Aspergillus terreus</i>	Decaying Acacia wood and industrial water effluent	CMC agar supplemented with chloramphenicol	Morphological characteristics and microscopic aspects	Bekele et al. 2015
<i>Aspergillus niger</i> IMMIS1	soil	CMC agar	Morphological characteristics and molecular characterization	Imran et al. 2018
<i>Aspergillus niger</i> VTCC-F021	Culture collection	CMC agar	28S rDNA sequencing	Pham et al. 2012
<i>Aspergillus terreus</i>	Soil	Sabouraud dextrose agar supplemented with filter paper strip	ITS 18S rDNA region sequencing	El-Baroty et al. 2019
<i>Aspergillus terreus</i> strain AKM-F3	Soil	CMC agar supplemented with Congo red	28S rDNA sequencing	Maharana and Ray 2015
<i>Peniophora</i> sp. NDVN01	Culture collection	CMC agar	18S rRNA gene sequencing	Trinh et al. 2013
<i>Thermoascus aurantiacus</i> RBB-1	Paper pulp recycling mill's soil	Potato dextrose agar (PDA) supplemented with CMC	ITS rDNA region sequencing	Dave et al. 2015
<i>Trichoderma harzianum</i> strain HZN11	Soil	PDA supplemented with CMC	18S rDNA sequencing	Bagewadi et al. 2016
<i>Trichoderma atroviride</i>	Soil	CMC agar	Morphological aspects and microscopic features	Bairagi 2016
<i>Trichoderma viride</i>	Culture collection	Lignocellulosic substrate agar	Morphological characteristics and microscopic features	Iqbal et al. 2011

ITS internal transcribed spacer, CMC carboxymethylcellulose

Bagewadi et al. 2016; Bairagi 2016). Similarly, Kirk et al. (2002) and Iqbal et al. (2011) reported that most of the cellulases that are commercialized are produced from *Trichoderma* and *Aspergillus* species. *Peniophora* sp. (Trinh et al. 2013) and *Thermoascus* sp. (Dave et al. 2015) were also reported as producers of the detergent fungal cellulases (Table 1).

Cost-effective and easily available growth supporting substrates are used to produce detergent compatible fungal cellulases. For instance, agricultural wastes, corn stalks, and sugarcane bagasse were utilized by *Aspergillus* species to produce detergent-compatible cellulases (Pham et al. 2012; Maharana and Ray 2015; Imran et al. 2018; El-Baroty et al. 2019). Sweet sorghum bagasse, vegetable waste, and wheat straw were cost-effective substances to produce detergent fungal cellulases in significant amounts (Iqbal et al. 2011; Ahmed et al. 2016; Bagewadi et al. 2016; Bairagi 2016). Dave et al. (2015) use a readily available cheap *Jatropha* deoiled seed cake to produce the cellulase from *Thermoascus aurantiacus* RBB-1. Therefore, the utilization of cellulosic material residues for fungal cellulases makes the production inexpensive and the pollution of the environment is monitored and decreased (Phitsuwan et al. 2013; Saxena and Singh 2014).

Cellulases production by fungal species is carried in solid-state fermentation (SSF) or submerged fermentation (SmF) and both strategies are generally cost-effective (Zhang and Zhang 2013). Indeed, some researchers prefer to use the SSF due to its simple process, functions under static conditions, minimal water output, high productivity, foam absence, cheap substrate utilization, less secretion of waste products, and low energy requirement; while others prefer SmF due to its simplicity of sterilization, simple enzyme recovery, heat and mass transfer, and controlled physicochemical and nutritional conditions (Bairagi 2016; Ahmed and Bibi 2018). Some detergent compatible fungal cellulases were produced under SSF. For instance, the detergent-compatible cellulases of *Trichoderma* species were produced with SSF (Iqbal et al. 2011; Ahmed et al. 2016; Bairagi 2016). *T. aurantiacus* RBB-1 produced a detergent-compatible cellulase under SSF with the response surface methodology carried out with Box-Behnken design (Dave et al. 2012). The detergent-compatible cellulases were also produced under SmF with *Aspergillus* species (Nguyen and Quyen 2010; El-Hadi et al. 2014; Bekele et al. 2015; Imran et al. 2018; El-Baroty et al. 2019). SSF or SmF is carried out sometimes at low temperature to reduce energy utilization and to reduce the contamination exposures. For example, the detergent-compatible cellulase of *A. terreus* strain AKM-F3 was conducted at 15 °C and energy consumption was enormously reduced (Maharana and Ray 2015). More efforts are still needed to find the optimal parameters for cellulases production by fungi with improved quality to minimize the cost.

Fungal co-culturing was found to be beneficial because the co-produced enzymes are produced in important amounts in

both solid and submerged fermentations. For instance, *Trichoderma viride* and *Aspergillus niger* produced cellulases in a significant quantity with waste paper (Juwaied et al. 2010) or wheat bran as substrate (Ikram-ul-Haq et al. 2005). Similarly, Jayant et al. (2011) screened *A. niger* and *Penicillium chrysogenum* that secreted cellulases in high amounts under SSF with waste paper. The co-culture of *Penicillium* and *Cladosporium* species was able to produce four hydrolases utilized in the detergent industry. These are cellulase, amylase, protease, and lipase (Abe et al. 2015). Two yeasts, viz., *Tetracladium* sp. and *M. gelida*, were co-cultured and produced high amounts of cellulases and amylases using a medium containing CMC or starch as a carbon source (Carrasco et al. 2016). *T. viride* and *A. niger*, after co-culturing, secreted cellulase and xylanase maximally (compared to monocultures) when wheat straw and sugarcane bagasse were used as substrates (Irfan et al. 2013). *Emericella nidulans* AUMC 5687 was able to concomitantly produce cellulase, xylanase, and pectinase in significant amounts (Moubasher et al. 2016). The co-culturing of fungi leads to a higher amount of cellulases and other enzymes production than corresponding monocultures. The process is environmentally friendly and cost-effective.

Optimization of nutritional and physicochemical factors for significant fungal cellulases production

The nutritional (like C and N sources) and physicochemical (inoculum size, pH, etc.) factors are the vital parameters affecting detergent-compatible cellulases. They have to be optimized for any fungus-producing the enzyme.

Initial pH of cultivation medium effect on detergent fungal cellulases production

Among physicochemical parameters, the initial pH of the cultivation medium is crucial because it induces morphological changes leading to detergent enzyme production (Casalheira and Queiroz 1999; Niyonzima and More 2013a). The initial pH of *Aspergillus* (Nguyen and Quyen 2010; Pham et al. 2012; Bekele et al. 2015; Imran et al. 2018), *Thermoascus* (Dave et al. 2015), and *Trichoderma* (Iqbal et al. 2011; Ahmed et al. 2016; Bagewadi et al. 2016; Bairagi 2016) species producing detergent-compatible cellulases are in the 4.5 to 6.0 range (Table 2). However, the initial neutral pH was also observed for cellulase production by *Aspergillus hortai* (El-Hadi et al. 2014) and *Peniophora* sp. NDVN01 (Trinh et al. 2013). A low pH of 3.0 was also recorded for cellulase secretion by *A. terreus* (El-Baroty et al. 2019) (Table 2). After optimum pH, a decrease in bacterial or fungal detergent enzyme was noticed. This could be attributed to the alteration of enzyme structure (Niyonzima and More 2013b; Bairagi 2016). The lower pHs below optimum were also found to

Table 2 Optimum physicochemical and nutritional conditions for fungi producing detergent-compatible cellulases

Microorganism	pH	T(°C)	Agitation (rpm)	Inoculum size (%)	Incubation time (days)	Good C source	Preferred N source	Reference
<i>Aspergillus terreus</i>	5	30	0	ns	4	CMC	Ammonium nitrate	Bekele et al. 2015
<i>Aspergillus hortai</i>	7	37	150	10	4	Lactose	Yeast extract	El-Hadi et al. 2014
<i>Aspergillus niger</i> IMMIS1	4.8	30	0	ns	7	CMC	Urea, yeast extract, and proteose peptone	Imran et al. 2018
<i>Aspergillus niger</i> VTCC-F021	4.5	37	200	ns	5	Sugarcane bagasse	Soybean	Pham et al. 2012
<i>Aspergillus oryzae</i> VTCC-F045	5.5	28	200	ns	5	Lactose	Soybean and ammonium sulfate	Nguyen and Quyen 2010
<i>Aspergillus terreus</i>	3	35	0	5	5	Microcrystalline cellulose	Peptone	El-Baroty et al. 2019
<i>Aspergillus terreus</i> strain AKM-F3	5	15	0	10	5	Lactose	Sodium nitrate	Maharana and Ray 2015
<i>Peniophora</i> sp. NDVN01	7	28	200	ns	7	Pulp	Ammonium monohydrogen phosphate	Trinh et al. 2013
<i>Thermoascus aurantiacus</i> RBB-1	4.6	50	0	ns	6	Jatropha deoiled seed cake	Jatropha deoiled seed cake	Dave et al. 2015
<i>Trichoderma harzianum</i> strain HZN11	6	35	0	7	7	Lactose	Ammonium sulfate and proteose peptone	Bagewadi et al. 2016
<i>Trichoderma atroviride</i>	6	30	180	ns	5	Sucrose	Yeast extract	Bairagi 2016
<i>Trichoderma harzianum</i>	5.5	35	180	10	7	Lignocellulosic substrate	Ammonium hydrogen sulfate	Ahmed et al. 2016
<i>Trichoderma viride</i>	5.5	45	0	10	7	Lignocellulosic substrate	Ammonium sulfate	Iqbal et al. 2011

ns not specified

reduce detergent-compatible enzyme activity because the growth of fungi gets inhibited (Niyonzima et al. 2013; Maharana and Ray 2015).

Influence of incubation temperature on fungal cellulases production

The temperature was reported as a factor influencing the enzyme production including detergent-compatible cellulases (Karmakar and Ray 2011; Niyonzima 2018). *Aspergillus* species (Pham et al. 2012; El-Hadi et al. 2014; Bekele et al. 2015; Imran et al. 2018; El-Baroty et al. 2019) and *Trichoderma* species (Ahmed et al. 2016; Bagewadi et al. 2016; Bairagi 2016) that secrete detergent-compatible cellulases are in the range of 30–37 °C. However, a low incubation temperature of 15 and 28 °C was observed for detergent-compatible cellulase production by *A. terreus* strain AKM-F3 (Maharana and Ray 2015) and *Peniophora* sp. NDVN01 (Trinh et al. 2013), respectively. The optimal temperatures for cellulases production by *Trichoderma viride* (Iqbal et al. 2011) and *T. aurantiacus* RBB-1 (Dave et al. 2015) noticed were 45 and 50 °C, respectively (Table 2). The reduction in detergent enzyme secretion after optimum temperature may occur due to the inhibition of microbial growth and thermal inactivation (Niyonzima and More 2014d; Maharana and Ray 2015).

Influence of shaking on fungal cellulases secretion

The fermentation flasks are normally agitated at moderate rpm to favor maximum enzyme production (Kuhad et al. 2011). Most of the fungal cellulases are produced under static conditions (Iqbal et al. 2011; Dave et al. 2015; Bekele et al. 2015; Maharana and Ray 2015; Bagewadi et al. 2016; Imran et al. 2018; El-Baroty et al. 2019). However, when shaken, 150–200 rpm agitation range is considered. For instance, 150 rpm was optimum for cellulase secretion by *Aspergillus hortai* (El-Hadi et al. 2014). Ahmed et al. (2016) and Bairagi (2016) reported 180 rpm as the best shaking condition for cellulases secretion by *Trichoderma* species. *Aspergillus* species (Nguyen and Quyen 2010; Pham et al. 2012) and *Peniophora* sp. NDVN01 (Trinh et al. 2013) produce cellulases at the maximum level when agitated at 200 rpm (Table 2). Indeed, when cultivation flasks are shaken, the nutrients are well mixed and available to all bacterial or fungal cells, booting enzyme production. The shaking also maintains the aeration at optimum.

Influence of inoculum size on fungal cellulases secretion

The inoculum level is one of the important process parameters to be investigated when producing detergent enzymes during fermentation (Gaur and Tiwari 2015). Fungi producing cellulases are usually inoculated with 10% (v/v) (Iqbal et al. 2011;

El-Hadi et al. 2014; Maharana and Ray 2015; Ahmed et al. 2016). However, 5 and 7% were seen as the best inoculum levels for detergent-compatible cellulases production by *A. terreus* (El-Baroty et al. 2019) and *Trichoderma harzianum* strain HZN11 (Bagewadi et al. 2016), respectively. Lower inoculation size leads to lower fungal growth owing to less conidial cells formation, whereas a higher inoculum concentration after optimum decreases detergent enzyme activity. This decrease can be attributed to the abundant fungal growth leading to fungal cell autolysis because of the nutritional imbalance. The decrease could also be attributed to the initial lag phase increase or depletion of important C and N sources or oxygen transfer (El-Hadi et al. 2014; Maharana and Ray 2015; Niyonzima 2019).

Influence of incubation time on fungal cellulases secretion

The incubation period has to be short to make fermentation inexpensive (Olama et al. 1993). Most fungi produce detergent-compatible cellulases from 5 to 7 days (Nguyen and Quyen 2010; Iqbal et al. 2011; Pham et al. 2012; Trinh et al. 2013; Dave et al. 2015; Maharana and Ray 2015; Ahmed et al. 2016; Bagewadi et al. 2016; Bairagi 2016; Imran et al. 2018; El-Baroty et al. 2019). However, a low incubation period of 4 days was recorded for cellulase secretion by *Aspergillus* species (El-Hadi et al. 2014; Bekele et al. 2015). A decrease in detergent enzyme production was observed after the optimal incubation period. This may be ascribed to the lack or decrease of important nutritional parameters in the production medium or poor oxygen transfer for fermentation carried out at 0 rpm (Maharana and Ray 2015; Bairagi 2016). The different incubation period reported may be attributed to the difference in the C sources bioavailability (Niyonzima and More 2014d; El-Baroty et al. 2019).

Influence of carbon sources on fungal cellulases secretion

A good choice of carbon sources is required since they have a vital role in the metabolism of a cell, and thus the detergent enzymes synthesis (Saha 2004; Bairagi 2016). Various C sources were reported to produce detergent in significant amounts (Table 2). For example, CMC and microcrystalline cellulose were C sources of choice for cellulase production by *Aspergillus* species (Bekele et al. 2015; Imran et al. 2018; El-Baroty et al. 2019). Low-cost substances such as sugarcane bagasse (Pham et al. 2012), pulp (Trinh et al. 2013), *Jatropha* deoiled seed cake (Dave et al. 2015), and lignocellulosic substrate (Iqbal et al. 2011; Ahmed et al. 2016) were also reported to produce fungal cellulases. Disaccharides such as lactose (El-Hadi et al. 2014; Bagewadi et al. 2016) and sucrose (Bairagi 2016) were also the best C sources. Indeed, when the lactose is used as a carbon source and an inducer, increase in enzyme activity is observed as the disaccharide penetration

Table 3 Purification steps for detergent fungal cellulases

Fungal species	Partial purification	Total purification	Specific activity (U / m g protein)	Fold increase	Yield (%)	Reference
<i>Aspergillus niger</i> IMMIS1	40–80% ammonium sulfate	Sephadex G-100 gel filtration	388	6.9	1.63	Imran et al. 2018
<i>Aspergillus niger</i> VTCC-F021	70% ammonium sulfate	Sephadex G100 gel filtration chromatography	14.122	2.09	18.4	Pham et al. 2012
<i>Aspergillus oryzae</i> VTCC-F045	65% ammonium sulfate saturation	Sephadex G100 gel filtration chromatography	40.36	2.6	6.0	Nguyen and Quyen 2010
<i>Aspergillus terreus</i> strain AKM-F3	40–95% ammonium sulfate	ns	75.279	3.464	23.925	Maharana and Ray 2015
<i>Pentophora</i> sp. NDVN01	90% ammonium sulfate	Bio-Gel P-100 and Sephadex G-75 gel filtration	163.8	2.8	3.6	Trinh et al. 2013
<i>Thermoascus aurantiacus</i> RBB-1	40% ammonium sulfate	DEAE cellulose DE-52 resin DE-52 ion-exchange, and Bio-Gel P-100 size exclusion chromatography	44.9	6.6	13.3	Dave et al. 2015
<i>Trichoderma harzianum</i> strain HZN11	70% ammonium sulfate	DEAE-Sepharose ion exchange followed by Sephadex G-100 gel filtration chromatography	66.25	33.12	5.9	Bagewadi et al. 2016
<i>Trichoderma harzianum</i>	50–80% ammonium sulfate	Gel filtration chromatography with the help of Sephadex G-100	101.05	1.83	2.0	Ahmed et al. 2016
<i>Trichoderma viride</i>	50–80% ammonium sulfate	Gel filtration chromatography with the help of Sephadex G-100	105	2.33	2.11	Iqbal et al. 2011

ns not studied

Table 4 Aspects of detergent compatible fungal cellulases

Fungal species	MW (kDa)	pH stability	T (°C)	Temperature stability	Metal ions	Specific reagent	Reference
<i>Aspergillus niger</i> IMMIS1	71	4.5	35	nd	nd	nd	Imran et al. 2018
<i>Aspergillus niger</i> VTCC-F021	31	> 60% residual activity from pH 5.0 to 6.0	55	> 80% stable up to 50 °C	Stimulated by 5-mM K ⁺ , Cu ²⁺ , and Fe ²⁺ at 5 mM; no effect with Co ²⁺ ; moderate inhibition with Ag ⁺ and Ca ²⁺ ; and strong inhibition with Zn ²⁺ , Ni ²⁺ , and Mn ²⁺	Activated by EDTA at 5 mM	Pham et al. 2012
<i>Aspergillus oryzae</i> VTCC-F045	36	5.5 60% residual activity range for 6 h	55	60% stable at up to 55 °C for 6 h	No effect with 4 mM K ⁺ , Ni ²⁺ , Cu ²⁺ , Mn ²⁺ ; slight inhibition with 4 mM Ca ²⁺ , Co ²⁺ , Fe ²⁺ , Zn ²⁺ , and Ag ⁺	No effect with 4-mM EDTA	Nguyen and Quyen 2010
<i>Aspergillus terreus</i> strain AKM-F3	55	4 90% stable in pH 5.0–8.0 range	15	80% stable at 35 °C	More stable in Ca ²⁺ and Mn ²⁺ ; 67.84 to 88.20% residual activity with Fe ³⁺ , Zn ²⁺ , Hg ²⁺ , Mg ²⁺ , Cu ²⁺ , Ba ²⁺ , Li ⁺ , Na ⁺ , and K ⁺	Slight inhibition with EDTA	Maharana and Ray 2015
<i>Peniophora</i> sp. NDVN01	32	4.5 Maximum stability in the pH range of 3.5–5.5 with an overnight residual activity of 77%	60	Stable at up to 42 °C with an overnight residual activity of 80%	Activated by Ni ²⁺ ; no inhibition with Ca ²⁺ and Zn ²⁺ ; slight inhibition with K ⁺ and Ba ²⁺ ; moderate inhibition with Na ⁺ , Fe ²⁺ , Mn ²⁺ , and Mg ²⁺ ; and complete inhibition with Ag ⁺ and Cu ²⁺	Stimulated by 2-ME; moderate inhibition with EDTA	Trinh et al. 2013
<i>Thermoascus aurantiacus</i> RBB-1	35	4 71% residual activity for 1 h	70	100% stable at 70 °C and 90% stable for 1 h at 80 °C	nd	nd	Dave et al. 2015
<i>Trichoderma harzianum</i> strain HZN11	55	5.5 Stable from pH 4.5 to 6.0 with 77% activity after 3 h	60	80% residual activity at 50 °C after 3 h	Stimulated by Ca ²⁺ , Mg ²⁺ , Co ²⁺ , Fe ²⁺ , and Mn ²⁺ ; inhibited by Hg ²⁺ , Cd ²⁺ , Zn ²⁺ , and Pb ²⁺	Activated by DTT, 2-ME, and urea; inhibited by NBS, p-CMB, PMSF, IAA, DMSO, 1,10-phenanthroline	Bagewadi et al. 2016
<i>Trichoderma harzianum</i>	43	6 Optimal stability from pH 5.0 to 8.0	50	Stable from 30 to 55 °C	Activated by Co ²⁺ and Mn ²⁺ ; inhibited by Hg ²⁺	Inhibited by EDTA	Ahmed et al. 2016
<i>Trichoderma viride</i>	58	8 Optimal stability from pH 5.0 to 8.0	55	Stable from 30 to 60 °C	Activated by Co ²⁺ and Mn ²⁺ ; inhibited by Hg ²⁺	Inhibited by EDTA	Iqbal et al. 2011

nd not determined

through the cell membrane is favored (El-Hadi et al. 2014). The glucose was found in many cases to decrease fungal growth and thus enzyme activity due to carbon catabolite repression (Ahmed and Bibi 2018; Niyonzima 2019).

Influence of nitrogen sources on detergent-compatible cellulases production by fungi

Preferred N sources were reported to be different for the cellulases secretion by fungi. Some fungal species used inorganic sources like ammonium nitrate (Bekele et al. 2015), sodium nitrate (Maharana and Ray 2015), ammonium monohydrogen phosphate (Trinh et al. 2013), and ammonium hydrogen sulfate (Ahmed et al. 2016) or organic sources such as yeast extract (El-Hadi et al. 2014; Bairagi 2016), soybean (Pham et al. 2012), and peptone (El-Baroty et al. 2019). The combination of nitrogen and organic nitrogen sources is sometimes considered to produce cellulase in an important amount. For instance, soybean and ammonium sulfate were mixed to produce cellulase in huge quantity from *A. oryzae* VTCC-F045 (Nguyen and Quyen 2010), whereas ammonium sulfate and protease peptone were combined to over-secrete cellulase from *T. harzianum* strain HZN11 in higher amounts (Bagewadi et al. 2016). A mixture of nitrogen organic sources was also taken into account for cellulase production by *Aspergillus niger* IMMIS1 (Imran et al. 2018). In some cases, a substance serves as both N and C source. Dave et al. (2015) utilized Jatropa deoiled seed cake as both C and N sources for detergent-compatible cellulase production by *T. aurantiacus* RBB-1. Sometimes, amino acids are supplemented to the production medium to increase cellulase secretion. For instance, aspartate acts as an inducer and boosts the detergent-compatible cellulase production from *A. terreus* strain AKM-F3 (Maharana and Ray 2015). A decrease in cellulase activity noticed after optimum could be ascribed to N source repression.

Purification of detergent-compatible cellulases produced by fungi

For partial purification of detergent-compatible enzymes, salts like ammonium sulfate and organic solvents like acetone and ethanol are used (Niyonzima and More 2014c). All the detergent-compatible fungal cellulases were precipitated by ammonium sulfate with saturation varying from 40 to 95%. The fungal cellulases need not to be in pure for detergent application. For example, the cellulase of *A. terreus* strain AKM-F3 was utilized in detergent formulation after only precipitation step (Maharana and Ray 2015). The fungal cellulases were mainly purified by gel filtration chromatography with Sephadex G-100 column packing materials (Nguyen and Quyen 2010; Iqbal et al. 2011; Pham et al. 2012; Ahmed et al. 2016; Imran et al. 2018). A combination of chromatographic

methods was also reported. For example, DEAE cellulose DE-52 resin ion-exchange chromatography followed by Biogel P-100 size exclusion chromatography was the desired chromatographical method to purify the cellulase of *T. aurantiacus* RBB-1 (Dave et al. 2015). The cellulase obtained from *T. harzianum* strain HZN11 was purified with DEAE-Sepharose ion exchange and Sephadex G-100 gel filtration chromatography (Bagewadi et al. 2016). Trinh et al. (2013) use the Bio-Gel P-100 and Sephadex G-75 gel filtration chromatography procedure to totally purify the detergent-compatible cellulase of *Peniophora* sp. NDVN01. The specific activity of cellulases from fungi is in the range of 40–160 U/mg protein with fold increase varying from 2.0 to 33.0 and with a recovery yield of 1.5–24% range (Nguyen and Quyen 2010; Iqbal et al. 2011; Trinh et al. 2013; Dave et al. 2015; Maharana and Ray 2015; Ahmed et al. 2016; Bagewadi et al. 2016). The lower and higher specific activity of 14.122 and 388 U/mg protein were also observed for *A. niger* VTCC-F021 (Pham et al. 2012) and *A. niger* IMMIS1 (Imran et al. 2018), respectively (Table 3).

Properties of detergent compatible fungal cellulases

Molecular weight of fungal cellulases

Due to genomic composition and differences, molecular weights of detergent-compatible enzymes were reported to be different. The molecular weights of cellulases purified from *Aspergillus* (Nguyen and Quyen 2010; Pham et al. 2012; Maharana and Ray 2015; Imran et al. 2018), *Peniophora* (Trinh et al. 2013), *Thermoascus* (Dave et al. 2015), and *Trichoderma* (Iqbal et al. 2011; Ahmed et al. 2016; Bagewadi et al. 2016) species are in the 31–71 kDa range. All these detergent-compatible cellulases were found to be monomeric proteins. Similar ranges were reported for enzymes used in the detergent preparations like 22–80 kDa for detergent bacterial cellulases (Niyonzima 2018), 30–94.5 kDa for detergent bacterial amylases (Niyonzima and More 2014a), and 16.1–60 kDa for detergent bacterial and fungal lipases (Niyonzima and More 2015b). Table 4 shows the characteristics of detergent fungal cellulases.

Influence of pH on fungal cellulases activity and stability

The enzyme to be used in the detergent industry has to be stable broadly in the alkaline region. The acidic pH in the range of 4.0–6.0 was observed for most of the fungal cellulases (Nguyen and Quyen 2010; Pham et al. 2012; Trinh et al. 2013; Dave et al. 2015; Maharana and Ray 2015; Ahmed et al. 2016; Bagewadi et al. 2016; Imran et al. 2018). However, 8.0 was the optimum pH of cellulase from *T. viride* (Iqbal et al. 2011). Although the optimum pH of fungal cellulases is in the acidic region, they are also stable in the alkaline region and

thus can be used in detergent formulations. For instance, the cellulase of *A. terreus* strain AKM-F3 was 90% stable in the pH 5.0–8.0 range (Maharana and Ray 2015). Similarly, 71% residual activity at pH 8.0 for 1 h was noticed for the cellulase obtained from *T. aurantiacus* RBB-1 (Dave et al. 2015). Iqbal et al. (2011) and Ahmed et al. (2016) purified detergent-compatible cellulases from *Trichoderma* species that had stability in the pH range of 5.0 to 8.0.

Celluzyme® obtained from *Humicola* sp. functions actively in the pH and temperature ranges of 4–10 and 25–70 °C, respectively. Similarly, Carezyme® from the same species has a similar temperature range, but a different pH application range of 5–10.5. Both commercial enzymes were developed by Novo Nordisk (Bagsvaerd, Denmark) (Olsen and Falholt 1998). The different optimum pH observed for these detergent enzymes may be ascribed to the variability between their genetic materials (Li et al. 2008; Niyonzima and More 2014c; Iqbal et al. 2011). The decline in enzyme activity observed after the optimum pH could be due to enzyme 3D structure alteration owing to action of the H⁺ or OH⁻ in the cellulase active site (Niyonzima and More 2014b; Maharana and Ray 2015).

Influence of temperature on fungal cellulases activity and stability

The optimum temperature observed for fungal cellulases varies from 35 to 60 °C (Nguyen and Quyen 2010; Iqbal et al. 2011; Pham et al. 2012; Trinh et al. 2013; Ahmed et al. 2016; Bagewadi et al. 2016; Imran et al. 2018). However, a low temperature of 15 °C was observed for a cellulase purified from *A. terreus* strain AKM-F3 (Maharana and Ray 2015). A higher optimum temperature was also recorded for cellulase of *T. aurantiacus* RBB-1 (Dave et al. 2015). All these fungal cellulases are 60 to 100% stable in the specified range. The optimum temperature of 50 °C was observed for commercial cellulases that were active in the washing powder at pH 8.5–9.0 range (Uhlrig 1998; Behera

et al. 2017). Similarly, the range found was the one recorded by Ito et al. (1989) where the real washing conditions took place at pH 10.0 and 40 °C for 20 min. Thermostability is a vital aspect of alkaline cellulase to be used in the detergent industries. Some detergent fungal cellulases are stable at high temperatures. For instance, the cellulase of *T. aurantiacus* RBB-1 was 100% stable at 70 °C and 90% stable for 1 h at 80 °C (Dave et al. 2015). This was a good aspect of industrial enzyme as there is no enzyme thermal inactivation. But currently, due to the high cost of electricity, detergent-compatible cellulases functioning at low temperatures are preferred. The denaturation of fungal cellulases noticed after optimum temperature could be due to non-covalent bonds rupture, disrupting thus the 3D enzyme structure (Okoshi et al. 1990; Maharana and Ray 2015).

Influence of various cations on the fungal cellulases

The detergent fungal cellulases behave differently when reacting with cations. Indeed, the cellulase of *Aspergillus niger* VTCC-F021 was stimulated by Cu²⁺, K⁺, and Fe²⁺; moderately inhibited by Ag⁺ and Ca²⁺; strongly inhibited by Zn²⁺, Ni²⁺, and Mn²⁺; and no effect with Co²⁺ (Pham et al. 2012). Nguyen and Quyen (2010) purified the cellulase from *Aspergillus oryzae* VTCC-F045 that does not affect K⁺, Ni²⁺, Cu²⁺, and Mn²⁺; but it was slightly inhibited by Ca²⁺, Co²⁺, Fe²⁺, Zn²⁺, and Ag⁺. *A. terreus* strain AKM-F3 secretes a detergent-compatible cellulase that was stimulated by Ca²⁺ and Mn²⁺; and partially inhibited by Fe³⁺, Zn²⁺, Hg²⁺, Mg²⁺, Cu²⁺, Ba²⁺, Li⁺, Na⁺, and K⁺ (Maharana and Ray 2015). The cellulase from *Peniophora* sp. NDVN01 was activated by Ni²⁺; no inhibition with Ca²⁺ and Zn²⁺; but slight, moderate, and complete inhibitions were observed with K⁺ and Ba²⁺, Na⁺, Fe²⁺, Mn²⁺, and Mg²⁺, and Ag⁺ and Cu²⁺, respectively (Trinh et al. 2013). An activation by Ca²⁺, Co²⁺, Mg²⁺, Fe²⁺, and Mn²⁺ and an inhibition by Hg²⁺, Cd²⁺, Zn²⁺, and Pb²⁺ were observed for cellulase obtained from *T. harzianum* strain HZN11 (Bagewadi et al. 2016).

Table 5 Kinetic parameters for detergent compatible fungal cellulases

Microorganism	Substrate specificity	Km	Vmax	Reference
<i>Aspergillus niger</i> IMMIS1	CMC	0.54 mM	19 mM/min	Imran et al. 2018
<i>Aspergillus niger</i> VTCC-F021	CMC	8.5815 mg/ml	20.121 U/mg	Pham et al. 2012
<i>Aspergillus terreus</i> strain AKM-F3	CMC	0.37 mg/ml	24.63 U/mg	Maharana and Ray 2015
<i>Peniophora</i> sp. NDVN01	Barley β-glucan	5.9 mg/ml	9804 U/mg	Trinh et al. 2013
<i>Thermoascus aurantiacus</i> RBB-1	CMC	37 mg/ml	82.6 U/min/mg	Dave et al. 2015
<i>Trichoderma harzianum</i> strain HZN11	CMC	2.5 mg/ml	83.75 U/mg	Bagewadi et al. 2016
<i>Trichoderma harzianum</i>	CMC	63 μM	156 U/ml	Ahmed et al. 2016
<i>Trichoderma viride</i>	CMC	68 μM	148 U/ml	Iqbal et al. 2011

Table 6 Compatibility of different fungal cellulases in detergent ingredients

Fungal species	Ionic surfactant	Nonionic surfactants	Oxidizing and bleaching agents	Local detergent	Reference
<i>Aspergillus niger</i> IMMIS1	ns	ns	ns	Residual activity of 95.5% in Ariel, 90.16% in Surf excel, 89.26% in Express power, 88.6% in Sunlight, and 83.9% in Bright	Imran et al. 2018
<i>Aspergillus niger</i> VTCC-F021	50% stable in 0.5% (w/v) SDS	> 80%.residual activity in 0.5% (w/v) Tween-20, Tween-80, and Triton X-100	ns	ns	Pham et al. 2012
<i>Aspergillus oryzae</i> VTCC-F045	20% residual activity in 0.4% SDS	78, 42, 45% residual activity in 0.4% Tween-20, Tween-80, and Triton X-100	ns	ns	Nguyen and Quyen 2010
<i>Aspergillus terreus</i> strain AKM-F3	98% stable in 1% SDS, but inhibition with sodium deoxycholate	More than 80% stability in Triton X-100, Tween-20, and Tween-80	ns	ns	Maharana and Ray 2015
<i>Peniophora</i> sp. NDVN01	Total inhibition by 5% SDS	Activated by from 1 to 5% Tween-20, Tween-80, Triton X-100, and X-114	ns	ns	Trinh et al. 2013
<i>Thermoascus aurantiacus</i> RBB-1	84% stable in SDS for 1 h	ns	ns	60% of residual activity in Ariel, Rin, Wheel, and SurfExcel	Dave et al. 2015
<i>Trichoderma harzianum</i> strain HZN11	90% stable in 1% (w/v) SDS	More than 80% residual activity in 1% (v/v) Tween-20, -40, and -80, as well as Triton X-100	85% in 1% (w/v) sodium tetraborate, 64% in sodium perborate, 73% in sodium hypochlorite, and 40% in hydrogen peroxide	Residual activity of 68% in Tide, 71% in Ariel, and 76% in Surf excel at 1% (w/v)	Bagewadi et al. 2016
<i>Trichoderma harzianum</i>	Inhibited by 1 mM SDS	ns	ns	Stimulated by Bonus and SurfExcel; and stable in Wheel and Ariel	Ahmed et al. 2016
<i>Trichoderma viride</i>	Inhibited by 1 mM SDS	ns	ns	More stable in SurfExcel and stable in Wheel, Ariel, and Bright Total	Iqbal et al. 2011

ns not studied

Iqbal et al. (2011) and Ahmed et al. (2016) reported detergent-compatible cellulases from *Trichoderma* species that were activated by Co^{2+} and Mn^{2+} , and inhibited by Hg^{2+} . In most cases, Ni^{2+} , K^+ , and Ca^{2+} stimulated and Hg^{2+} inhibited the detergent fungal species. Indeed, the detergent fungal enzymes may require Ni^{2+} , K^+ , and Ca^{2+} for optimum activity and maximal stability; while Hg^{2+} may bind to some groups like carboxyl, thiol, and indole group present in the detergent enzyme active site and makes covalent bonds, thereby inhibiting enzyme (Trinh et al. 2013; Niyonzima and More 2014b; Maharana and Ray 2015; Niyonzima 2018).

Influence of specific reagents on fungal cellulases activity

The detergent-compatible cellulases purified from *Aspergillus* species are not inhibited by EDTA (Nguyen and Quyen 2010; Pham et al. 2012; Maharana and Ray 2015). They are therefore suitable for detergent use as stability in EDTA is needed. Indeed, the importance of EDTA in detergent preparation is that it improves soil removal by complexing the cations necessary for the hardness of water (Niyonzima and More 2015d). However, the inhibition was observed with cellulases of *Trichoderma* species (Iqbal et al. 2011; Ahmed et al. 2016) and thus not suitable for detergent industries. The decrease in detergent enzyme activity by EDTA may suggest the reaction between catalytic inorganic groups with EDTA, forming a complex which is inactive (Trinh et al. 2013; Niyonzima and More 2014c). 2-mercaptoethanol (2-ME) stimulated detergent-compatible cellulases of *Peniophora* sp. NDVN01 (Trinh et al. 2013) and *Trichoderma harzianum* strain HZN11 (Bagewadi et al. 2016). The stimulation of this cellulase shows that there is no thiol group in the active site (Niyonzima 2018). The cellulase of *T. harzianum* strain HZN11 was also activated by dithiothreitol (DTT) and urea. It was also inhibited by N-bromosuccinimide (NBS), p-chloromercuribenzoate (p-CMB), phenylmethylsulphonyl fluoride (PMSF), iodoacetamide (IAA), dimethyl sulfoxide (DMSO), and 1,10-phenanthroline. The inhibition of this detergent-compatible cellulase by IAA and p-CMB may suggest the presence of thiol functional groups. These thiol groups in the active site were confirmed by stimulation of the enzyme by DTT and 2-ME. Tryptophan may also be present in the active site as the cellulase is inhibited by NBS (Bagewadi et al. 2016).

Broad substrate specificity and kinetic parameters of detergent fungal cellulases

CMC was the best substrate for detergent fungal cellulases in most cases compared to other substrates tested (Iqbal et al. 2011; Pham et al. 2012; Dave et al. 2015; Maharana and Ray 2015; Ahmed et al. 2016; Imran et al. 2018). For instance, Bagewadi et al. (2016) obtained a cellulase from *T. harzianum*

strain HZN11 that was active against CMC, followed by filter paper and Avicel. However, the cellulase from *Peniophora* sp. NDVN01 has a higher specificity with barley β -glucan. The activity was four times compared to the one of CMC, but the cellulase was inactive against Avicel, locust bean gum, and birchwood xylan. It was therefore an endoglucanase (Trinh et al. 2013). The best substrate was also CMC for detergent bacterial cellulases (Niyonzima 2018). Kinetic parameters of fungal cellulases utilized in the detergent preparations are in Table 5. V_{max} and K_{m} are two main kinetic constants utilized to study detergent enzymes. K_{m} found for most of fungal cellulases is in the 0.015–8 mg/ml range (Iqbal et al. 2011; Pham et al. 2012; Maharana and Ray 2015; Ahmed et al. 2016; Bagewadi et al. 2016; Imran et al. 2018). Low K_{m} shows the highest cellulase activity for the CMC substrate. However, the cellulase of *T. aurantiacus* RBB-1 has relatively higher K_{m} of 37 mg/ml compared to others (Dave et al. 2015). The difference in K_{m} and V_{max} observed between detergent enzymes from fungal species may be ascribed to their genetic variabilities (Iqbal et al. 2011; Niyonzima and More 2014b).

Influence of oxidizing and surfactant agents on fungal cellulases

Sodium dodecyl sulfate (SDS) is regarded as a component of detergent. The cellulases obtained from *Aspergillus* (Pham et al. 2012; Maharana and Ray 2015), *Thermoascus* (Dave et al. 2015), and *Trichoderma* species (Bagewadi et al. 2016) were stable in SDS and thus useful in detergent manufacturing. However, an inhibition was observed for some fungal cellulases (Nguyen and Quyen 2010; Iqbal et al. 2011; Trinh et al. 2013; Ahmed et al. 2016) (Table 6). SDS is importantly used in detergent formulation to react with positive ions necessary for water hardness (Niyonzima and More 2014c).

Nonionic surfactants are also important components of liquid and powder detergents. Most of the obtained cellulases were stable in Tween-20, Tween-80, and Triton X-100. Indeed, Pham et al. (2012) purified cellulase from *A. niger* VTCC-F021 that possessed significant activity in Tween-20, Tween-80, and Triton X-100. *A. terreus* strain AKM-F3 produced a detergent-compatible cellulase with good stability in Triton X-100, Tween-80, and Tween-20 (Maharana and Ray 2015). The cellulase from *Peniophora* sp. NDVN01 was stimulated by Tween-20, Tween-80, Triton X-100, and X-114 (Trinh et al. 2013). Tween 80, Triton X-100, Tween 20, and Tween 40 had no negative influence on the cellulase obtained from *T. harzianum* strain HZN11 (Bagewadi et al. 2016). Surfactants activate enzyme activities by favoring the permeability of surface-bound enzymes (Bairagi 2016). The cellulase of *A. niger* VTCC-F021 was partially inhibited with a residual activity of 78, 42, and 45% in Tween 20, Tween 80,

and Triton X-100, respectively (Nguyen and Quyen 2010) (Table 6).

Fungal detergent-compatible cellulases have to be stable in oxidizing and bleaching agents. For example, *T. strain HZN11* secreted a detergent-compatible cellulase with important activity in sodium tetraborate, sodium perborate, sodium hypochlorite, and hydrogen peroxide (Bagewadi et al. 2016). A small amount of oxidizing agents, surfactants, and bleaching agents have to be used as these can cause pollution to the environment. Bajpai and Tyagi (2007) proposed a detergent composition as follows: 0.004% (w/v) detergent enzymes, 0.14% (w/v) bleaching and oxidizing agents, and 0.02 and 0.03% (w/v) for nonionic and ionic surfactants, respectively.

Stability of fungal cellulases in local detergents

The fungal cellulases have to possess 100% stability in detergent components. The fungal cellulases were checked if they can resist in presence of commercial detergent components. An important stability was observed for cellulase from *A. niger* IMMIS1 in Ariel, Surf excel, Express power, Sunlight, and Bright (Imran et al. 2018). Dave et al. (2015) purified cellulase from *T. aurantiacus* RBB-1 that had a significant activity in Ariel, Rin, Wheel, and Surf Excel. Tide, Ariel, and Surf excel did not affect the cellulase activity of *T. harzianum* strain HZN11 (Bagewadi et al. 2016). The cellulase of *T. harzianum* was stimulated by Bonus and Surf Excel, and possessed maximum activity in in Wheel, and Ariel (Ahmed et al. 2016), whereas the cellulase of *Trichoderma viride* was activated by only Surf Excel and had 100% stability in Wheel, Ariel, and Bright Total (Iqbal et al. 2011) (Table 6).

Low velocities were necessary for BaCel5 and HiCel45 cellulases to remove the stains from fabrics at pH 10.0 and 40 °C for 20 min when used in liquid detergents. In these conditions, the softness and brightness of the fabrics are restored by using low dosages, and thus higher rates are not required for depilling as they can have destructive effects like loss of fabric strength (Caparrós et al. 2012). Celluzyme® and Carezyme® are also used in the powder and liquid detergents formulation to maintain the fabric softness, smoothness, and brightness. They are therefore known as color clarification cellulases (Olsen and Falholt 1998). Cellulase AP30K purified from *Aspergillus niger* supplied by Amano enzyme and Cellulase TRL obtained from *Trichoderma longibrachiatum* and *Trichoderma reesei* commercialized by Solvay Enzymes (Elkhart, IN) showed good properties to remove the soils from fabrics (Begum and Absar 2009). The detergent-compatible cellulase of *Trichoderma harzianum* strain HZN11 exhibited extraordinary storage stability of 60 days with 87% residual activity (Bagewadi et al. 2016).

In laundry detergent, 1–3% (w/w) of cellulase is incorporated to allow the soil removal and to give fabrics color brightening, softening, and whiteness maintenance. However, proteases, lipases, and amylases are added to both types of detergents in the range of 0.2–2% for laundry and 0.5–3% (w/w) for dish-wash detergents (Olsen and Falholt 1998). The fungal cellulase obtained from *Trichoderma harzianum* was able to remove ink-stain from a white cloth and the quality of the fabric was improved (Ahmed et al. 2016). In detergent industries, the fungal cellulase is used because it removes loose fibrils resulted from mechanical abrasion when used at a low dose. This selective degradation of only fibrils may be ascribed to the enzyme lower crystallinity enzyme and specificity in its action (Boisset et al. 1997). Ito et al. (1989) suggested that cellulases act by weakening cellulose amorphous areas where the soils prefer to bind.

Genetic engineering of fungal cellulases used in detergents

Genetic engineering and directed evolution can be used to enhance the activity and stability, to develop more efficient, and to reduce the cost of detergent-compatible cellulases production. For instance, the stability of Cel45 cellulase from *Humicola insolens* during detergent formulation was observed after the protein engineering step. Indeed, the surfactant C12-LAS inactivated the cellulase obtained from *H. insolens* Cel45 in detergent preparation. Site-directed mutagenesis was utilized to create mutations in the Cel45 crystal structure and cellulase with important stability in the detergent industry was obtained. The stability was shown by a significant resistance towards mechanical agitation while washing (Otzen et al. 1999). A high level of cellulase production was noticed after transferring the gene *stce1* into the fungus *H. insolens* (Kuhad et al. 2011). Wang et al. (2014) expressed cellulase genes in *T. reesei* based on promoters and terminators. The bio-engineered alkaline cellulase was able to remove fibrils on the fabric surface, a process known as bio-stoning. Thus, research in genetic and protein engineering has to continue to get inexpensive detergent-compatible cellulases with desirable properties, as the production cost is still high.

Like other enzymes, cellulase secretion may be regulated by artificial or native transcription factors at the transcription level. Various filamentous fungi were reported to possess many transcription regulators. For example, artificial and natural transcription factors were manipulated in *T. reesei* to overproduce cellulases (Zhao et al. 2018). Krull et al. (2013) reported maximum cellulase production through pellet morphology engineering. The physicochemical parameters controlled may include shaking, temperature, inoculation concentration, pH, medium composition, and culture mode. Genome editing at the chromatin level was also utilized to develop improved fungal strains that over-produce cellulases (Zhao

et al. 2018). Therefore, different and various genetic engineering methods can be applied to produce fungal cellulases for detergency by modifying the fungal genetic material. Indeed, genetic engineering can help fungi to cost-effectively produce significant amounts of detergent compatible-cellulases.

Conclusion

Detergent compatible fungal cellulases were reviewed in term of production, characteristics, and stability in the presence of other detergent components. Although important researches have been conducted on isolation, identification, production, purification, and characterization of detergent compatible fungal cellulases, future investigations should mostly focus on gene/protein engineering of these cellulases to overproduce stable and inexpensive fungal cellulases. The use of low-cost substrates including agricultural residues and spent byproducts has to continue to be exploited because the process is environmentally friendly and makes the enzyme production cost-effective. More efforts are still needed to fully understand the properties of detergent compatible fungal cellulases. The continual discovery of detergent enzymes with desirable aspects at low cost will allow the formulation of strong detergents that can remove all tough soil types. The eminent researchers from academia and industry have to work together to achieve all these goals.

Acknowledgments The author thank INES Ruhengeri for encouragement.

Compliance with ethical standards

Conflict of interest The author declares no conflict of interest.

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