

REVIEW OF LITERATURE

Organic wastes from renewable forest and agricultural residues comprise cellulose, hemicellulose and lignin in an average ratio of 4:3:3 (Brauns and Brauns, 1960). Lignocellulosic wastes are generated through forestry, agricultural practices and industrial processes, particularly from agro-allied industries such as breweries, paper-pulp, textile and timber industries. These wastes generally accumulate in the environment thereby causing pollution problem (Abu *et al.*, 2000).

Energy conversion, utilization and access underlie many of the great challenges of our time, including those associated with sustainability, environmental quality, security and poverty. Biotechnology could give rise to important new energy conversion processes. Resources for biological conversion of energy to forms useful to humanity include majorly the plant biomass. Among forms of plant biomass, lignocellulosic biomass is particularly well suited for energy applications because of its large scale availability, low cost and environmentally benign production (Larry *et al.*, 1982).

Utilization of lignocelluloses as a substrate for ethanol production has a barrier in its complex structure, which resists degradation lignocelluloses is composed of three main fractions cellulose (45%), hemicellulose (30%) and lignin (25%) (Tomati *et al.*, 1995). Cellulose is the most abundant polymer and can be hydrolyzed chemically or enzymatically. Lignin is the most abundant aromatic polymer in nature, is a macromolecule of phenolic character (Schoemaker *et al.*, 1985) and binds cellulose and hemicelluloses making it resistant to degradation.

Among the annual plants hard woods and softwoods contain 20-25% and 7-12% xylan respectively (Whistler and Richards 1970). The complete hydrolysis of xylan requires the combined action of various enzymes such as endoxylanase and several accessory enzymes to hydrolyse substituted xylan residues by endwise attack of xylooligosaccharide (Wong *et al.*, 1988).

Hemicellulose is a composite of different non-cellulosic polysaccharide, xylan being the major polysaccharide, glucan and mannan being present to a lesser content.

It is a polymer of xylose containing β -1,4 xylosidic linkages (Biswas *et al.*, 1986). Hemicellulose is composed mainly of xylan that constitutes about 20-40% of total plant biomass. It is a linear polymer of β -D Xylopyranosyl unit linked by (1 to 4) glycosidic bonds (Ninawe *et al.*, 2008).

The hemicelluloses are those polysaccharides, soluble in alkali and are associated as a cementing matrix between cellulose and lignin. The principal monomers present in most of the hemicelluloses are D-xylose, D-mannose D-galactose and L-arabinose. The main heteropolymers are xylan, mannan, galactan and arabinan, xylan contains D-xylose as monomeric unit and traces of L-arabinose, galactan consists of D-galactose and mannan is made up of D-mannose units, while arabinan is composed of L-arabinose.

Many microorganisms including bacterial strains of *Acidobacterium*, *Aeromonas*, *Bacillus*, *Bacteroides*, *Cellulomonas*, *Microbacterium*, *Paenibacillus*, *Ruminococcus* and *Streptomyces*, *Thermoanaerobacterium* and yeast strains of *Aureobasidium*, *Cryptococcus* and the fungal strains of *Acrophilophora*, *Aspergillus*, *Cephalosporium*, *Fusarium*, *Geotrichum*, *Paecilomyces*, *Penicillium*, *Thermomyces* and *Trichoderma* are known to produce different type of xylanases and the nature of the enzymes varies between these different organisms (Beg *et al.*, 2001; Abdelwahed *et al.*, 2011).

The xylan degrading enzymes, xylanases, have attracted increasing attention in biotechnological research during the past decade largely because of their potential application in cellulose pulp bleaching (Viikari *et al.*, 1986), degradation of plant cell wall materials (Omar *et al.*, 2008). The manufacture of food, bread and drinks, textiles, bleaching of cellulose pulp, ethanol and xylitol production, biofuel production and Xylooligosaccharide production (Subramaniyan and Prema 2002; Salupi *et al.*, 2015).

Only a few xylanases exhibit stability under high temperature and alkaline pH conditions including xylanases produced from *Bacillus* which are comparatively thermostable and can tolerate a wide pH range (Haki *et al.*, 2003) but the cost of production and the low yield of these enzymes are the two major problems for industrial applications (Techapun *et al.*, 2003).

Cheaper hemicellulosic substrate like corn cob, wheat bran, rice bran, rice straw, corn stalk and bagasse have also been found to be most suitable for the production of xylanases (Haltrich *et al.*, 1994). Among fungi, the maximum activity reported is 3350 IU / ml in *Trichoderma reesei* (Haapala *et al.*, 1994). The highest xylanase activity reported from a bacterial host was 36633 IU/mg in *Bacillus subtilis* (Guo *et al.*, 2012).

Many of the xylanases are produced by alkalophilic organisms such as *Bacillus* sp (Okazaki *et al.*, 1984). Members of *Bacillus* produce large variety of extracellular enzymes of which xylanases have particularly significant industrial importance (Annamalai *et al.*, 2009)

2.1 ENZYMATIC ACTION ON XYLAN IN THE LIGNOCELLULOSIC BIOMASS

2.1.1 XYLANOCCURRENCE AND DISTRIBUTION:

Schulze (1891) first introduced the term “hemicellulose” for the fractions isolated or extracted from plant materials with dilute alkali. Hemicelluloses include xylan, mannan, galactan and arabinan as the main heteropolymers. The classification of these hemicelluloses fractions depends on the types of sugar moieties present.

Xylan is the most abundant non-cellulosic polysaccharide present in hard woods and annual plants, accounts for 20 to 35% of total dry weight in tropical plant biomass. In temperate soft woods, xylans are less abundant and may comprise about 8% of total dry weight.

Xylan is found mainly in the secondary cell wall and is considered to be forming an inter phase between lignin and other polysaccharides. Xylans are linear homopolymers that contain D-xylose monomer linked through β -1-4 glycosyl bonds (Srinivasan *et al.*, 1999).

The primary walls of monocotyledons plants include as a major hemicelluloses, an arabinoxylan with rather more glucuronic acid. The primary walls of dicotyledons have small amounts of glucuronoarabinoxylan whereas the secondary wall contains glucuroxylan.

Xylan is the major hemicellulose in hardwood from angiosperms, but is less abundant in softwood from gymnosperms it accounts for approximately 15-30% and 7-12% of the total dry weight, respectively (Whistler and Richards 1970; Wong *et al.*, 1988).

Homoxylans, on the other hand, consist exclusively of xylosyl residues. Xylan isolated from marine algae (Barry and Dhillon, 1940). This type of xylan is not widespread in nature and has been isolated from esparto grass (Chanda *et al.*, 1950).

2.1.2 THE BACKBONE OF XYLAN:

The main chain of xylan is composed of β -(1-4) linked β -xylapyranose residues. The presence of β -(1-4) linkages in between two adjacent xylose residues in xylan was reported by Aspinall (1959) and Chanda *et al.*, 1950. The presence of β -(1-4) glycosidic linkages was demonstrated by Jayme & Satee (1942) and Whistler (1950).

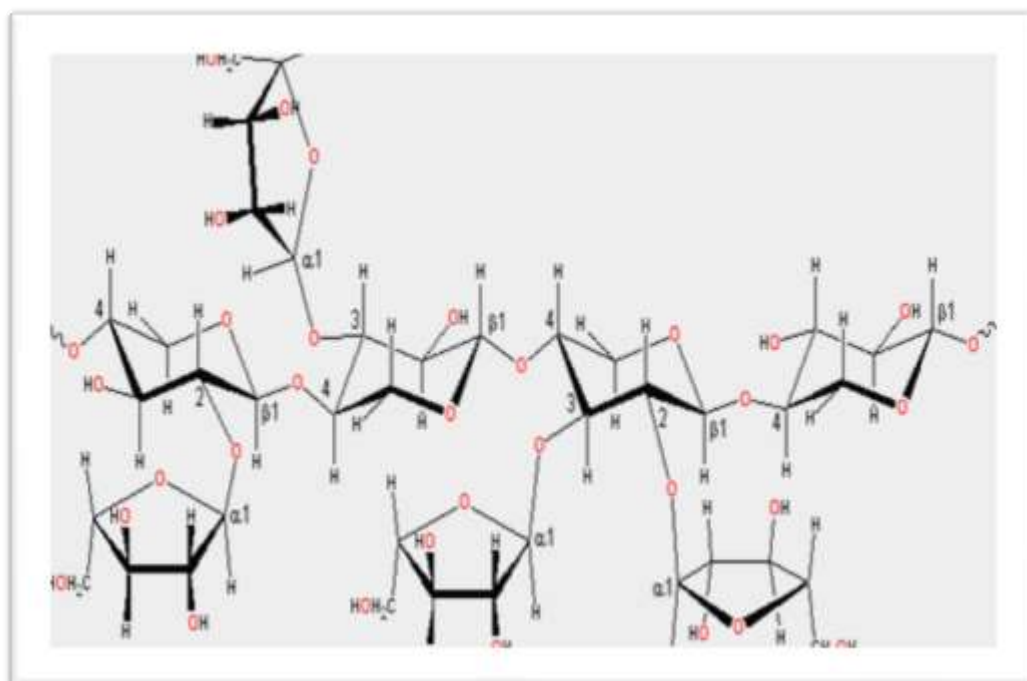
2.1.3 XYLAN PROPERTIES:

- a) Deacetylated xylans are insoluble in water, but soluble in alkaline solutions and are easily hydrolysed with acids.
- b) Acetylated xylans can be extracted by hot water and more soluble in water.
- c) Xylan solutions show high negative optical rotation.
- d) Xylan solutions show high negative optical ranging from $[\alpha]_D^{20}$ -78.2 to 109.5°C (Whistler 1950)
- e) Acetylated xylans are easily degraded by microbial enzymes.

2.1.4 STRUCTURE OF XYLAN:

The structure of xylan found in cell walls of plants can differ greatly depending on their origin but they always contain a β -1, 4 linked D-xylose backbone (Ebringerova and Heinze, 2000).

Structural unit of Xylan (Ebringerova and Heinze, 2000)



Arabinose is connected to the backbone of xylan via α -1,2 or 1,3 linkage either as single residue or as short side chains. Glucuronic acid and its 4-O methyl ether are attached to the xylan backbone via an α -1,2 linkage, whereas aromatic feruloyl and p-coumaroyl residues have so far been found attached only to O-5 terminal arabinose residues (Saulnier *et al.*, 1995). As a consequence of all these features the xylans form very heterogeneous groups of polysaccharides (Bajpai, 1997).

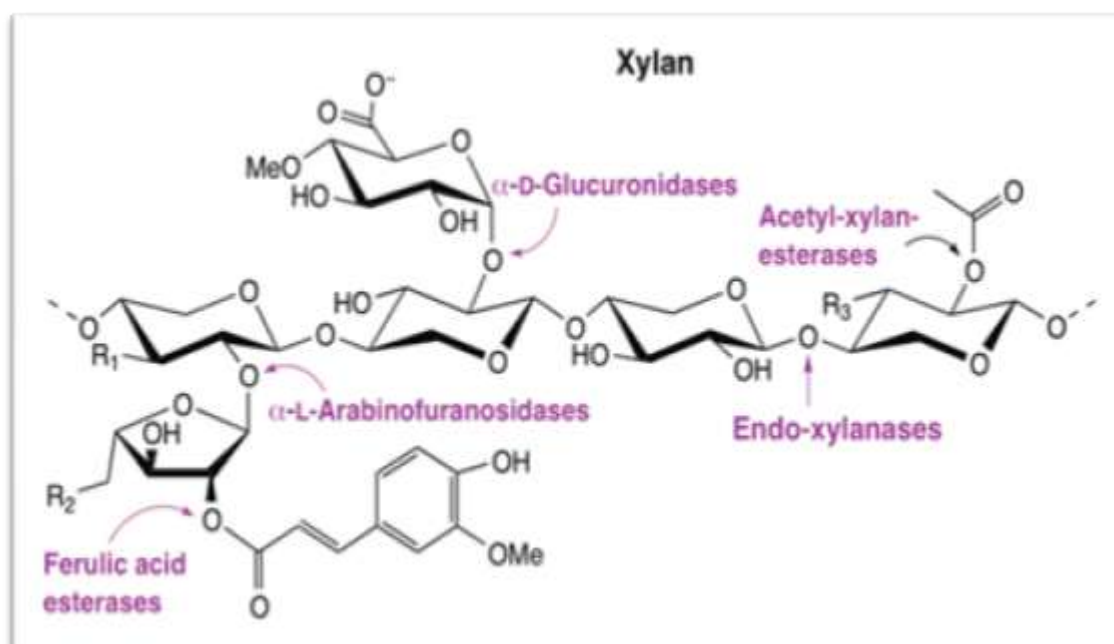
2.1.5 BIODEGRADATION OF XYLAN:

The total biodegradation of xylan requires endo β ,1-4 xylanase, β -xylosidase and several accessory enzymes such as α -arabino furanosidase, α -glucuronidase, acetyl xylan esterase, ferulic acid esterase and p-coumaric acid esterase which are necessary for hydrolyzing various substituted xylans. The following table indicates the list of enzymes involved in the degradation of xylan and their mode of action (De Boy *et al.*, 2008).

Table 2.1

Major Enzymes involved in the biodegradation of heteroxylan

Enzymes	Mode of action
Endo xylanase	Hydrolyzes mainly interior β ,1-4xylose linkags of the xylan backbone
Exo xylanase	Hydrolyzes β , 1-4 xylose linkages releasing xylobiose
β -xylosidase	Releases xylose from xylobiose and short chain xylooligosaccharides
α -arabinofuranosidase	Hydrolyzes terminal non reducing α -arabinofuranose from arabino xylans
α -glucuronidase	Releases glucuronic acid from glucuronoxylans
Acetyl acid esterase	Hydrolyzes feruloyl ester bonds in acetyl xylan
Ferulic acid esterase	Hydrolyzes acetyl ester bands in xylan
P-coumaric acid esterase	Hydrolyzes p-coumaryl ester bonds in xylans.

Biodegradation of xylan by enzymes (DeBoy *et al.*, 2008)

2.1.6 XYLANOSOME

Xylanosomes are discrete, multifunctional, multienzyme complexes found on the surface of several microorganisms (Sunna and Antranikian, 1997). These complexes play an important role in the degradation of hemicelluloses.

2.1.7 TYPES OF XYLAN:

There are two types of hemicelluloses, the acetylated xylan of hardwood and arabinoxylan of softwood. The degree of polymerization of hardwood xylans (150-200) is higher than that of softwoods (70-130) (Timell, 1967).

Hardwood xylan is typically O-acetyl 4-O methyl glucuronic xylan with approximately 10% xylose units substituted with α 1-2 linked 4-O methyl glucuronic acid side chain and 70% of xylose residues are acetylated at the C2 or C3 position. Acetylation occurs more frequently at the C3 and double acetylation of a D-xylose unit has also been reported (Bouveng, 1967). The presence of acetyl groups makes the xylan significantly soluble in water. It constitutes about 15-30% of the cell wall content.

Soft wood xylan is commonly arabinoxylan in which 10% of xylose units are substituted with α -2-3 linked arabinofuranose residues. It consists about 7-10% of the cell wall content. (Whistler, 1970 and Biely, 1985).

Table 2.2

Methods for pretreatment of lignocellulosic biomass (Saha, 2003)

Methods	Example
Thermo-Mechanical Autohydrolysis	Grinding, Milling, Shearing, Extruder, Steam Pressure, Steam Explosion
Acid Treatment	Dilute & Concentrated H ₂ SO ₄ , HCl
Alkali Treatment	Sodium hydroxide, Ammonia, Alkaline hydrogen Peroxide
Organic Solvents Treatment	Methanol, Ethanol, Butanol, Phenol

2.1.8 EXTRACTION OF XYLAN:

Alkali was also used for the pretreatment of lignocellulosic biomass and its action depends upon the lignin content present in the biomass (Fan *et al.*, 1987; McMillan, 1994). Panbangred *et al.* (1983) achieved maximum xylan extraction from agrowastes using 3% NaOH. Hemicelluloses are extracted effectively from lignified tissues of grasses and woody plants by alkali treatment. However, partial extraction of hemicelluloses from plants is achieved by hot/cold water or dilute alkali. Generally 4 to 10% KOH or NaOH is used.

Prolonged treatment of rye flour and barley husk with 7% NaOH caused a 20% decrease in the molecular weight of these polymers (Aspinall *et al.*, 1959). The fractions extracted dilute alkali contained low molecular weight xylan fractions, whereas the concentrated alkali treatment selectively removed the higher molecular weight fractions. Hagglund *et al.* (1956) used Dimethyl Sulphoxide (DMSO) for hemicelluloses extraction from wood holocellulose. Xylan and arabinoxylan is extracted from kraft pulps by removing the glucoside uronic acid residues with Ba(OH)₂ (Hamilton *et al.*, 1958). Xylan extraction has also been investigated by microwave treatment (Wanitwattanaramulg *et al.*, 2012).

Table 2.3

Carbohydrate Compositions of various xylans(Li *et al.*,2000)

Carbohydrate residues (Mole %)					
Name of the xylans	Birch Wood Xylan	Oat Spelt Xylan	Barley Husk Xylan	Larch Wood Xylan	Yellow Poplar Xylan
Arabinose		22.3	15.0		
Rhamnose			0.9		4.2
Xylose	94.1	52.5	61.4	47.5	31.0
Glucose	1.4	15.7	7.1	26.5	38.0

Galactose	4.5	9.5	14.2		10.0
Mannose				26.0	9.0
Glucuronic Acid			0.8		
Galacturonic Acid			0.6		6.5
N-acetyl glucosamine					1.3

Table 2.4

Xylan extraction from different authors

Substrate	Author & Year	Alkali	Acid
Sugarcane bagasse	Bocchini <i>et al.</i> , 2005		0.5 ml of H ₂ SO ₄
Sugarcane bagasse	Irfan <i>et al.</i> , 2014	2% H ₂ SO ₄ , 2.5% KOH, 3% H ₂ O ₂	
Sugarcane bagasse	Mahalakshmi and Jayalakshmi, 2016	5% NaOH	1% H ₂ SO ₄
Corn cob	Ebringerova <i>et al.</i> , 1998	5% NaOH	
Corn cob	Yang <i>et al.</i> , 2005		1.0 g/l H ₂ SO ₄
Corn cob	Richana <i>et al.</i> , 2007	1% NaOCl	
Corn husk, Sugarcane bagasse, Corn cob, Rice straw, Rice bran	Tachaapaikoon <i>et al.</i> , 2006	1N NaOH	
Lignified tissue of grasses & woody plants	Bastawde, 1992	4% to 10% KOH (or) NaOH	
Barley straw	Rezaeian <i>et al.</i> , 2005	3 ml of NaOH	

Both hardwood and softwood xylans have a reducing end group constituting of rhamnosyl, galactouronosyl and xylosyl residues (Anderson, 1983).

The chemical composition of hard and soft woods are given in Table 2.5

Table 2.5

Comparative account of cell wall components in soft and hard wood (Anderson, 1983)

Chemical	Softwood (%)	Hard wood (%)
Cellulose	37-42	42-51
Glucomannan	15-20	1-3
Xylan	4-6	20-30
Other Polysaccharides	3-5	2-4
Lignin	27-32	21-26
Extratives	2-5	1-4

2.2 MICROBIAL XYLANASES:

Xylanase (EC.3.2.1.8: endo- β -1-4-D-xylanase) is mainly responsible for the hydrolysis of xylan with β -1-4-xylanolytic linkages. Enzymes are biological catalysts produced by all living things. The enzyme named xylanase deconstructs plant structural material by breaking down hemicelluloses, a major component of the plant cell wall. Plant cell walls are necessary to prevent dehydration and maintain physical integrity. They also act as a physical barrier to attack by plant pathogens. In nature, some plant consumers or pathogens use xylanase to digest or attack plants. Many microorganisms produce xylanase, but mammals do not. Some herbivorous insects

and crustaceans also produce xylanase consumption as well as low environment pollution (Sweeney *et al.*, 2012).

Microbial enzymes are the preferred choice for conversion of lignocellulosic wastes into useful products and they provide high specificity, low energy or chemical

Microbial xylanases are the preferred catalysts for xylan hydrolysis due to their

- High specificity
- Mild reaction conditions
- Negligible substrate loss
- Side product generation

However, the cost of enzymatic hydrolysis of biomass is one of the factors limiting the economic feasibility of the process. The production of xylanases must therefore be improved by finding more potent fungal or bacterial strains or by inducing mutant strains to excrete greater amounts of enzymes.

2.2.1 OCCURRENCE OF XYLANASES:

Xylanases are widely distributed. They occur in both prokaryotes and eukaryotes (Dekker and Richards 1976) and have been demonstrated in higher eukaryotes, including protozoa, insects, snails and germinating plant seeds. Amongst the prokaryotes, bacteria and cyanobacteria from marine environments produce xylanases (Dekker, 1985), Extracellular and intracellular xylanases from various bacterial and fungal source have been studied extensively. Intracellular xylanase occur in rumen bacteria and protozoa (Dekker and Richards, 1976).

Occurrence of Multiple xylanases in Micro organisms:-

Multiple xylanases have been reported in numerous micro organisms (Dekker, 1985). Three different xylanases have been purified from the culture filtrate of *Clostridium stercorarium* and in *streptomyces sp* (Marui *et al.*, 1985).

2.2.2 CLASSIFICATION OF XYLANASE:

On the basis of molecular weight, pH, activity profile and specificity xylanases have been broadly divided into two families, Family F(GH11) and Family G (GH10) xylanases of hydrolases (Thomas, 1996).

Family G or GH 10 family have a low molecular mass with a pI between 8-9.5 and the Family F or GH 11 family have a high molecular mass and lower pI values.

2.2.3 TYPES OF XYLANASES:

Three different xylanases are involved in xylan degradation (Dekker, 1985)

a) **Endo- β (1-4)-D-xylanase [β -(1-4) D-xylan, xylano Hydrolyse] [EC 3.2.1.8]:**

These enzyme acts randomly on xylan to produce large amounts of xylo-oligosacchaides of various chain lengths. There are four types:

- i. **Non-arabinose Liberating Endoxylanases:** These cannot act on L-arabinosyl initiated branch points at β -(1-4)linkages and produce only xylobiose and xylose as the major end products. These enzymes can breakdown xylo oligosaccharides as small as xylobiose.
- ii. **Non-arabinose Liberating Endoxylanases:** These cannot cleave branch points at α -(1-2)and α -(1-3) and produce mainly xylo-oligosaccharides larger than xylobiose. These endoxylanases have no action on xylotriose and xylobiose.
- iii. **Arabinose Liberating Endoxylanases:** These can cleave the xylan chain at the branch points an produce mainly xylobiose, xylose and arabinose.
- iv. **Arabinose Liberating Endoxylanases:** These can hydrolyze the branch points and produce intermediate size xylooligosaccharides and arabinose.

b) **Exo- β - (1-4)D-xylanase [β -(1-4)D-xylan xylohydrolase]:** These enzymes removethe single xylose units from the non-reducing end of the xylan chain.

- c) **β -xylosidase or xylobiase [EC 3.2.2.37]**: These enzymes hydrolyze disaccharides like xylobiose and the higher xylo-oligosaccharide with decreasing specific affinity.

2.2.4 XYLANASE ASSAY METHOD

Methods used for the assay of xylanase are reported by many workers. Most of them report xylanase activities based on the release of reducing sugars from partially soluble xylan substrates (Tan *et al.*, 1985).

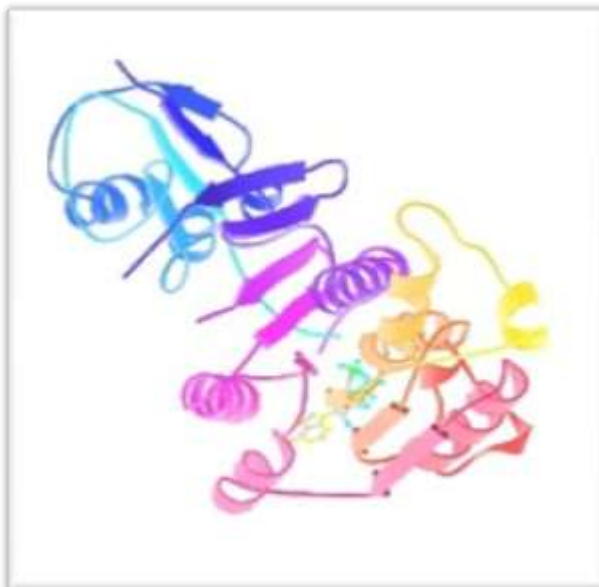
Sugar detection by the DNS methods (Di Nitro Salicylic acid) was chosen by many workers rather than Somogyi-Nelson (SN) method. This is because SN method is known to give a lower result than DNS (Breull and Saddler, 1985), Cup-plate clearance zone method (Gairola and Powell, 1971), Remazol Brilliant Blue (RBB) xylan method (Biely, 1985), Fluorescence –based method (Motta *et al.*, 2013).

2.2.5 CELLULASE –FREE XYLANASES:

Most microorganisms produce both xylanases and cellulose. Several strategies such as selective in activation of accompanying cellulose by mercurial compounds, bulk scale purification recombinant DNA technology to selectively express xylanase gene in non-cellulolytic host have met with limited success especially from the view point of commercial feasibility.

However it was in the mid-eighties that cellulose free xylanase was from a Sclerotial Actinomycete *Chainia*, isolated from the desert sands of Rajasthan, India. (Srinivasan *et al.*, 1999), in *Bacillus* (Subramaniyan and Prema, 2000), in *Coprinellus disseminates* (Agnihotri *et al.*, 2010). Certain strains of *Aspergillus* are the potential source of cellulose-free xylanase (Biely, 1985) and in *Aspergillus niger* DX-23 (Desai and Iyer, 2016)

Structure of Bacterial xylanase



2.2.6 BACTERIAL XYLANASES:

Bacterial xylanases hydrolyse xylan to xylotriase and higher oligosaccharides. Intensive investigations have been performed with xylanolytic enzymes derived from bacteria, both aerobic and anaerobic. Very few bacterial xylanases have been well characterized and most have been found to be endoxylanases producing xylobiose and xylotriase as the main end products. Some of the bacterial xylanases are tabulated.

Table 2.6

List of xylanolytic bacteria studied by various authors

Bacteria	References
<i>Bacillus pumilus</i>	Panbangred <i>et al.</i> , 1983; Moriya <i>et al.</i> , 2005; Battan <i>et al.</i> , 2007; Kapoor <i>et al.</i> , 2008 ; Bajaj <i>et al.</i> , 2012 ; Sethi <i>et al.</i> , 2013 ; Adhyaru <i>et al.</i> , 2014 ; Lalit kumar <i>et al.</i> , 2014.
<i>Bacillus flavothermus</i>	Sunna <i>et al.</i> , 1997
<i>Bacillus cereus</i>	Tremblay and Archibald, 1993

<i>Bacilluslicheniformis</i>	Archana and Satyanarayana, 1998; Damiano <i>et al.</i> , 2003.
<i>Bacillusstearothermophilus</i>	Fishman <i>et al.</i> , 1995
<i>Bacillus coagulans</i>	Heck <i>et al.</i> , 2005; Chauhan <i>et al.</i> , 2006
<i>Bacillus fibrisolvens</i>	Hespell <i>et al.</i> , 1987
<i>Bacillus halodurans</i>	Lin <i>et al.</i> , 2013; Prakash <i>et al.</i> , 2012
<i>Bacillus circulans</i>	Dhillon and Khanna 2000; Bocchini <i>et al.</i> , 2005 ; Pokhrel <i>et al.</i> , 2012
<i>Bacillus subtilis</i>	Bernier <i>et al.</i> , 1983; Panbangred <i>et al.</i> , 1983; Tarayre <i>et al.</i> , 2013; Irfan <i>et al.</i> , 2012; Sugumaran <i>et al.</i> , 2013; Ho <i>et al.</i> , 2015
<i>Bacillus firmis</i>	Chang <i>et al.</i> , 2004
<i>Bacillus safensis</i>	Rahmani <i>et al.</i> , 2014
<i>Bacillus atitudinis</i> DHN8	Adhyaru <i>et al.</i> , 2014
<i>Bacillus aerophilus</i> KGJ2	Gowdhaman <i>et al.</i> , 2014
<i>Bacillus lpuarvinder</i> st lpu 002	Kaur <i>et al.</i> , 2015
<i>Bacillus</i> sp	Gessesse and Gashe, 1997; Lopez <i>et al.</i> , 1998; Subramaniyan and Prema 2000; Senthil kumar <i>et al.</i> , 2005; Avcioglu <i>et al.</i> , 2005
<i>Butyrivibrio fibrisolvens</i>	Roger <i>et al.</i> , 1983; Bernier <i>et al.</i> , 1983
<i>Cellulomonas flavigenia</i>	Avalos <i>et al.</i> , 1996; Estrada <i>et al.</i> , 2002; Hernandez <i>et al.</i> , 2007
<i>Clostridium</i> sp	Marichamy and Mattiasson, 2005
<i>Clostridium absonum</i>	Swaroop Rani and Krishna Nand, 2000
<i>Clostridium thermolacticum</i>	Debeire <i>et al.</i> , 1990
<i>Cephalosporium stercoriarum</i>	Berenger <i>et al.</i> , 1985

<i>Caldocellumsaccharolyticum</i>	Luthi <i>et al.</i> , 1990
<i>Dictyoglomus</i> sp	Ratto <i>et al.</i> , 1994; Adamsen <i>et al.</i> , 1995
<i>Eschericia coli</i>	Liu <i>et al.</i> , 2003.
<i>Geobacillus thermoleovorans</i>	Verma and Satyanarayana, 2012
<i>Geobacillus atearothermophilus</i>	Wang <i>et al.</i> , 2013
<i>Geobacillus thermdentrificans</i>	Anand <i>et al.</i> , 2013
<i>Geobacillus stearothermophilus</i>	Bibi <i>et al.</i> , 2014
<i>Geobacillus</i> sp	Wu <i>et al.</i> , 2006
<i>Mesorhizzobium</i> sps	Sethi <i>et al.</i> , 2013
<i>Pseudomonas flaviginosa</i>	Penbroke <i>et al.</i> , 1992
<i>Pseudomonas</i> sp	Xu <i>et al.</i> , 2005
<i>Paenibacillus</i> sp	Harada <i>et al.</i> , 2008
<i>Paenibacillus terrae</i>	Song <i>et al.</i> , 2014
<i>Paenibacillus</i> sp XJ18	Yun <i>et al.</i> , 2015
<i>Streptomyces cyaneus</i>	Ninawe and Kuhad, 2005; Ninawe <i>et al.</i> , 2008
<i>Streptomyces</i> sp	Techapun <i>et al.</i> , 2001; Chungool <i>et al.</i> , 2006
<i>Strptomyces violaceroruber</i>	Khurana <i>et al.</i> , 2007
<i>Strptomyces olivaceoviridis</i>	Ding <i>et al.</i> , 2004; Ai <i>et al.</i> , 2005
<i>Staphylococcus aureus</i>	Iloduba <i>et al.</i> , 2016
<i>Staphylococcus</i> sp	Gupta <i>et al.</i> , 2000
<i>Thermusthermophilus</i>	Lyon <i>et al.</i> , 2000
<i>Thermotoga maritime</i>	Tan <i>et al.</i> , 2008

2.2.7 FUNGAL XYLANASES

Fungi are of special interest in the hydrolysis of cell wall polysaccharides because they produce high levels of extra cellular xylanase in the culture medium (Maheswari and Kamalam, 1985). Fungi are known to produce a good amount of β -xylosidase in addition to other enzymes occur in nature. Some of the common sources of fungal xylanases are given in the following Table 2.7

Table 2.7

List of xylanolytic fungi studied by various authors

Fungi	References
<i>Aspergillusniger</i>	Takenishi and Tsujisaka, 1975; Kim <i>et al.</i> ,1997; Tokuda <i>et al.</i> , 1997; Gawande and Kamat 1999; Haq <i>et al.</i> , 2002;Oxafor <i>et al.</i> , 2007;Soliman <i>et al.</i> , 2012; Sethi <i>et al.</i> , 2013; Desai and Iyer, 2016
<i>Aspergillusversicolor</i>	Jeya <i>et al.</i> , 2005
<i>Aspergillusterreus</i>	Gawande and Kamat, 1999
<i>Aspergillus oryzae</i>	Bailey and Viikari 1993; Sangeetha <i>et al.</i> , 2004
<i>Aspergillusfumigatus</i>	Bailey and Viikari, 1993; Lenartovicz <i>et al.</i> , 2002; Anthony <i>et al.</i> , 2003
<i>Aspergillusflavus</i>	Ruckmani and Rajendran, 2001
<i>Aspergillusawamori</i>	Botella <i>et al.</i> , 2007
<i>Arthrobacter sp</i>	Khandeparker and Bhosle, 2006
<i>Acremonium celluolyticus</i>	Kishishita <i>et al.</i> ,2014
<i>Acrophialophoranainiana</i>	Salles <i>et al.</i> , 2000; Cardoso <i>et al.</i> , 2003
<i>Coniochaetaligniaria</i>	Lopez <i>et al.</i> , 2007

<i>Fusariumavenaceum</i>	Sobezak and Urbanek, 1981
<i>Fusariumverticillioides</i>	Saha, 2001
<i>Geotrichumcandidum</i>	Rodionova <i>et al.</i> , 2000
<i>Melanocarpusalbomyces</i>	Roy <i>et al.</i> , 2003
<i>Neurosporacrassa</i>	Mishra <i>et al.</i> , 1984
<i>Penicilliumchrysogenum</i>	Okafor <i>et al.</i> , 2007
<i>Penicilliumoxalicum</i>	Li <i>et al.</i> , 2007
<i>Penicillium rolfsii</i>	Lee <i>et al.</i> , 2015
<i>Phanerochaetechrysosporium</i>	Nasser <i>et al.</i> , 1997; Dutt <i>et al.</i> , 2007
<i>Thermomyceslanuginosus</i>	Hoq <i>et al.</i> , 1995; Lin <i>et al.</i> , 1999; 2005; Sonia <i>et al.</i> , 2005; Stephens <i>et al.</i> , 2007
<i>Streptomyces</i> sp	Kusakabe <i>et al.</i> , 1983; Marui <i>et al.</i> , 1985; Lumba <i>et al.</i> , 1992; Flores <i>et al.</i> , 1997
<i>Streptomycesolivaceoviridis</i>	Ding <i>et al.</i> , 2004
<i>Streptomycescyaneus</i>	Ninawe and Kuhad, 2005
<i>Trichodermaharzianum</i>	Senior <i>et al.</i> , 1998; Silveria <i>et al.</i> , 1999
<i>Trichodermakoningii</i>	Li <i>et al.</i> , 2000
<i>Trichodermaviride</i>	Gomes <i>et al.</i> , 1992; Soliman <i>et al.</i> , 2012; Sethi <i>et al.</i> , 2013
<i>Thermoactinomycesthalophilus</i>	Kohli <i>et al.</i> , 2001
<i>Talaromycesbyssochlamydoides</i>	Yoshioka <i>et al.</i> , 1981

2.2.8 PRODUCTION OF XYLANASE BY LIGNOCELLULOSIC BIOMASSES

Agricultural by products that contain cellulose, hemicellulose and lignin could serve as inexpensive sources for xylanase production (Techapun *et al.*, 2003) Sugarcane baggase, wheat bran, rice bran, maize bran, gram bran, wheat straw, rice husk, rice straw, soy hull, sago hampas, grape vine trimmings dust, saw dust, corn cobs, coconut coirpith, banana waste, cassava waste, palm oil mill waste, paper pulp, sugar beet pulp, sweet sorghum pulp, apple pomace, peanut meal, rape seed cake, coconut oil cake, mustard oil cake, cassava flour, wheat flour, corn flour, steamed rice, stream pretreated willow, starch etc. According to Gomes *et al.* (2000) Cheaper hemicellulosic substrates namely cotton fibre, corn cob, wheat bran, paddy straw, paddy husk, sugarcane baggase, corn stalk, tamarind seed, saw dust and wheat straw can also be used as substrates for xylanase production. Among the substrates used, wheat straw served as a good substrate for xylanase production by *Cryptococcus adeliue*.

Wheat bran served as a good carbon source for xylanase production by in *Bacillus licheniformis* (Archana and Satyanarayana, 1998; Bajaj and Manhas, 2012). Orange peel supported maximum enzyme production by *Geobacillus stearothermophilus* (Bibi *et al.*, 2014), wheat straw enhanced maximum xylanase production in *Cryptococcus adeliue* (Gomes *et al.*, 2000), *Bacillus lpuarvinder* st lpu 002 (Kaur *et al.*, 2015). Rice bran supported maximum enzyme production in *Bacillus aerophilus* KGJ2 (Patel and Prajapati, 2014).

2.2.9 SUBSTRATES:

The most important factor influencing xylanase production or any enzyme production is the selection of a suitable substrate. Using commercial oat spelt xylan for xylanase production will be uneconomical compared to agricultural wastes. No single substrates works well with all organisms. Each organism prefers its own substrates for xylanase production.

Many workers have used various substrates for xylanase production. High extracellular xylanase was produced by cultures grown on hemicellulosic materials as substrate (Maheswari and Kamalam, 1985). Wheat bran was considered as the most

common and suitable metabolic substrate waste (Biswas *et al.*, 1986). Some of the agricultural residues which are used as substrates are tabulated in the Table 2.8.

Table 2.8

List showing cheaper lignocellulosic biomasses as substrates for xylanase production

Substrates	Organisms	References
Wheat bran	<i>Aspergillus niger</i>	Kana <i>et al.</i> , 2003; Stoilova <i>et al.</i> , 2007; Okafor <i>et al.</i> , 2007; Dobrev <i>et al.</i> , 2007; Irfan <i>et al.</i> , 2012; Sethi <i>et al.</i> , 2013
	<i>Aspergillus flavus</i>	Ruckmani and Rajendran, 2001
	<i>Aspergillus tamari</i>	Ferreira <i>et al.</i> , 1999
	<i>Aspergillus fischerii</i>	Senthil Kumar <i>et al.</i> , 2005
	<i>Aspergillus giganteus</i>	Coelho and Carmona, 2003
	<i>Aspergillus foetidus</i>	Chapla <i>et al.</i> , 2010
	<i>Arthrobacter sp</i>	Khande parkar and Bhosle, 2006
	<i>Fusarium oxysporum</i>	Panagiotou <i>et al.</i> , 2003
	<i>Trichoderma viride</i> IR05	Irfan <i>et al.</i> , 2014
	<i>Trichoderma longibrachiatum</i>	Azin <i>et al.</i> , 2007
	<i>Penicillium chrysogenum</i>	Okafor <i>et al.</i> , 2007
	<i>Streptomyces cyaneus</i>	Ninawe <i>et al.</i> , 2006
<i>Streptomyces sp</i>	Beg <i>et al.</i> , 2001; Bajaj <i>et al.</i> , 2010; Ghasemi <i>et al.</i> , 2014;	

		Sivakumar and Sharmal Kumar, 2016
	<i>Bacillus pumilus</i>	Kapoor <i>et al.</i> , 2008
	<i>Bacillus</i> sp	Azeri <i>et al.</i> , 2010
	<i>Bacillus mojavensis</i>	Sepahy <i>et al.</i> , 2011
	<i>Bacillus licheniformis</i>	Bajaj and Manhas, 2012
	<i>Geobacillus stearothermophilus</i>	Bibi <i>et al.</i> , 2014
Wheat flour	<i>Bacillus subtilis</i>	Sukumaran <i>et al.</i> , 2013
Wheat straw	<i>Aspergillus foetidus</i>	Chapla <i>et al.</i> , 2010
	<i>Trichoderma viride</i>	Gomes <i>et al.</i> , 1992; Goyal <i>et al.</i> , 2008
	<i>Trichoderma longibrachiatum</i>	Azin <i>et al.</i> , 2007
	<i>Streptomyces</i> sp	Lumba <i>et al.</i> , 1992
	<i>Bacillus circulans</i>	Dhillon and Khanna, 2000
	<i>Bacillus pumilus</i>	Kapoor <i>et al.</i> , 2008
	<i>Bacillus coagulans</i>	Chauhan <i>et al.</i> , 2006; Irfan <i>et al.</i> , 2012
Rice bran	<i>Bacillus</i> sp	Azeri <i>et al.</i> , 2010
	<i>Aspergillus niger</i>	Tokuda <i>et al.</i> , 1997
	<i>Aspergillus foetidus</i>	Chapla <i>et al.</i> , 2010
	<i>Arthrobacter</i> sp	Khande parkar and Bhosle, 2006
	<i>Thermoascus aurantiacus</i>	Santos <i>et al.</i> , 2003

	<i>Bacillus pumilus</i>	Poorna and Prema 2006
	<i>Bacillus licheniformis</i>	Bajaj and Manhas, 2012
	<i>Paenibacillus</i> sp	Harada <i>et al.</i> , 2006
Rice husk	<i>Trichoderma viride</i> IR05	Irfan <i>et al.</i> , 2014
	<i>Bacillus subtilis</i>	Irfan <i>et al.</i> , 2012
	<i>Bacillus pumilus</i>	Bajaj <i>et al.</i> , 2012
	<i>Bacillus licheniformis</i>	Bajaj and Manhas, 2012
Rice straw	<i>Aspergillus niger</i>	Kim <i>et al.</i> , 1997; Kana <i>et al.</i> , 2003
	<i>Aspergillus terreus</i>	Gawande and Kamat, 1999
	<i>Paenibacillus campinasensis</i> BL11	Ko <i>et al.</i> , 2010
	<i>Bacillus pumilus</i>	Poorna and Prema 2006
	<i>Bacillus circulans</i>	Heck <i>et al.</i> , 2006
	<i>Bacillus mojavensis</i>	Sepahy <i>et al.</i> , 2011
Sorghum straw	<i>Thermomyces lanuginosus</i>	Sonia <i>et al.</i> , 2005
Jowar straw	<i>Trichoderma viride</i>	Goyal <i>et al.</i> , 2008
Bajra straw	<i>Trichoderma viride</i>	Goyal <i>et al.</i> , 2008
Maize straw	<i>Trichoderma viride</i>	Goyal <i>et al.</i> , 2008
Barley straw	<i>Bacillus</i> sp	Gandarillas <i>et al.</i> , 2012
Barley husk	<i>Bacillus subtilis</i>	Ho <i>et al.</i> , 2015

Barley bran	<i>Aspergillus niger</i>	Soliman <i>et al.</i> , 2012
	<i>Trichoderma viride</i>	Soliman <i>et al.</i> , 2012
	<i>Bacillus mojavensis</i> A21	Haddar <i>et al.</i> , 2012
Sugarcane bagasse	<i>Aspergillus oryzae</i>	Sangeetha <i>et al.</i> , 2004
	<i>Trichoderma viride</i> IR05	Irfan <i>et al.</i> , 2014
	<i>Arthrobacter</i> sp	Khande parkar and Bhosle, 2006
	<i>Humicola lanuginose</i>	Anand and Vithayathil, 1990
	<i>Streptomyces</i> sp	Flores <i>et al.</i> , 1997
	<i>Streptomycesolivaceoviridis</i>	Ding <i>et al.</i> , 2004
	<i>Bacillus circulans</i>	Bocchini <i>et al.</i> , 2005
	<i>Bacillus pumilus</i>	Poorna and Prema 2006; Bajaj <i>et al.</i> , 2012
Sugarcane pulp	<i>Bacillus mojavensis</i>	Sepahy <i>et al.</i> , 2011
	<i>Aspergillus niger</i>	Oxafor <i>et al.</i> , 2007
Cassava bagasse	<i>Penicilliumchrysogenum</i>	Okafor <i>et al.</i> , 2007
	<i>Aspergillus oryzae</i>	Sangeetha <i>et al.</i> , 2004
Corn cob	<i>Bacillus subtilis</i>	Sukumaran <i>et al.</i> , 2013
	<i>Aspergillus oryzae</i>	Sangeetha <i>et al.</i> , 2004; Irfan <i>et al.</i> , 2012
	<i>Trichoderma viride</i> IR05	Irfan <i>et al.</i> , 2014
	<i>Streptomyces</i> sp	Kusakabe <i>et al.</i> , 1983

	<i>Streptomycesolivaceoviridis</i>	Ding <i>et al.</i> , 2004
	<i>Thermomyces lanuginosus</i>	Hoq and Deckwer 1995
Corn flour	<i>Bacillus licheniformis</i>	Bajaj and Manhas, 2012
Corn fibre	<i>Fusarium verticillioides</i>	Saha, 2001
Corn husk	<i>Bacillus</i> sp	Tachaapaikoon <i>et al.</i> , 2006
Saw dust	<i>Aspergillus niger</i>	Okafor <i>et al.</i> , 2007
	<i>Penicilliumchrysogenum</i>	Okafor <i>et al.</i> , 2007
	<i>Bacillus pumilus</i>	Poorna and Prema 2006; Bajaj <i>et al.</i> , 2012
Soyabean fibre	<i>Bacillus circulans</i>	Heck <i>et al.</i> , 2006
Soybean hull	<i>Aspergillus terreus</i>	Gawande and Kamat, 1999
	<i>Aspergillus niger</i>	Gawande and Kamat, 1999
	<i>Bacillus circulans</i>	Heck <i>et al.</i> , 2006
Soya bean meal	<i>Trichoderma viride</i> IR05	Irfan <i>et al.</i> , 2014
	<i>Bacillus subtilis</i>	Irfan <i>et al.</i> , 2012
Sun flower meal	<i>Trichoderma viride</i> IR05	Irfan <i>et al.</i> , 2014
Gram husk	<i>Streptomycescyaneus</i>	Ninawe <i>et al.</i> , 2006
Black gram husk	<i>Streptomycescyaneus</i>	Ninawe <i>et al.</i> , 2006
Cotton seed husk	<i>Streptomycesolivaceoviridis</i>	Ding <i>et al.</i> , 2004
Lemon peel	<i>Streptomyces</i> sp	Flores <i>et al.</i> , 1997
<i>Cymbopoganmartin</i>	<i>Phanerochaete chrysosporium</i>	Dutt <i>et al.</i> , 2007

<i>i</i>		
Grape pomace	<i>Aspergillus awamori</i>	Carolina <i>et al.</i> , 2007
Coconut pith	<i>Bacillus pumilus</i>	Poorna and Prema 2006
Maize stalk	<i>Streptomycescyaneus</i>	Ninawe <i>et al.</i> , 2006
Jute fibre	<i>Trichoderma viride</i>	Gomes <i>et al.</i> , 1992
Jute sticks	<i>Trichoderma viride</i>	Gomes <i>et al.</i> , 1992
Oat bran	<i>Bacillus mojavensis</i>	Sepahy <i>et al.</i> , 2011
Oat hay	<i>Trichoderma viride</i>	Goyal <i>et al.</i> , 2008
Palm fiber	<i>Aspergillus terreus</i>	Lakshmi <i>et al.</i> , 2009
Bamboo leaves	<i>Mesorhizobium</i> sp	Sethi <i>et al.</i> , 2013
Peepal leaves	<i>Mesorhizobium</i> sp	Sethi <i>et al.</i> , 2013
Banana stem waste	<i>Mesorhizobium</i> sp	Sethi <i>et al.</i> , 2013
Banana Pseudostem	<i>Pleurotus sajor-caju</i>	Medeiros <i>et al.</i> , 2007
Barseem hay	<i>Trichoderma viride</i>	Goyal <i>et al.</i> , 2008
Crushed bajra	<i>Bacillus licheniformis</i>	Bajaj and Manhas, 2012
Tomato, Potato, Pepper residues	<i>Pleurotus ostreatus</i>	Morais <i>et al.</i> , 2005

2.3 CULTURAL CONDITIONS FOR XYLANASE PRODUCTION:

2.3.1 EFFECT OF pH ON XYLANASE PRODUCTION

Each micro organism requires an optimum pH for its growth and activity. The initial pH of the medium may influence many enzymatic systems and transport of enzymes across the cell membrane. pH of the cultural media is an important factor that influences xylanase production. The optimum pH for xylanase production was pH 4 to 4.9 in *Arachinotus* sp. (Rafi *et al.*, 1997), at pH 5 to 5.5 in *Penicillium janthinellum* (Palma *et al.*, 1996), at pH 6.5 in *Acrophialophora nainiana* (Cardoso and Filho, 2003), *Geobacillus stearothermophilus* (Bibi *et al.*, 2014), at pH 7 in *Bacillus altitudinus* DHN8 (Adhyaru *et al.*, 2014), at pH 8.6 in *Bacillus* sp. (Kaur *et al.*, 2015).

2.3.2 EFFECT OF TEMPERATURE ON XYLANASE PRODUCTION

The optimum temperature for endoxylanase production by bacteria and fungal sources varies between 40°C to 60°C (Kulkarni *et al.*, 1999). The optimum temperature for xylanase production was 45°C in *Achromobacter xylosoxidans* (Mahalakshmi & Jayalakshmi 2016). In *Bacillus* sp. the enzyme activity was maximum at 40°C (Chi *et al.*, 2015), and 60°C in *Geobacillus thermodenitrificans* (Anandkumar and Satyanarayana, 2013).

2.3.3 EFFECT OF NITROGEN SOURCES ON XYLANASE PRODUCTION

Nitrogen sources have got profound influence on xylanase production, as it is the ultimate precursor for the protein biosynthesis. Besides the nitrogen sources can affect the pH of the medium, which in turn may influence the enzyme activity and stability. Yeast extract increased the enzyme production in *Bacillus pumilus* (Lalit kumar *et al.*, 2014) and *Bacillus* sp (Kaur *et al.*, 2015). Urea and peptone supported maximum xylanase yield in *Streptomyces* sp (Sivakumar & Sharmal kumar, 2016). Sodium nitrate supported maximum Xylanase production in *Achromobacter xylosoxidans* (Mahalakshmi and Jayalakshmi, 2016).

2.3.4 Effect of sugars on xylanase production

Oat spelt xylan enhanced maximum enzyme production in isolated fungal species of Amazon forest (Medeiros *et al.*, 2002). Glucose repressed xylanase production in *Bacillus Circulans* (Qureshy *et al.*, 2002). Xylose repressed xylanase production in *Bacillus* sp. (Gessesse & Gashe, 1997). Sucrose stimulate enzyme production in *Trichoderma harzianum* (Seyis and Aksoz, 2005).

2.3.5 EFFECT OF METAL IONS, DETERGENTS AND INHIBITORS ON XYLANASE ACTIVITY

The xylanase activity was stimulated by Fe^{2+} , Ni^{2+} , Cu^{2+} and DDT upto 60% and 100% inhibition in the presence of Co^{2+} , Hg^{2+} , Pb^{2+} , EDTA (Gupta *et al.*, 2000), EDTA and Hg^{2+} strongly inhibited xylanase activity in *Bacillis subtilis* (Annamalai *et al.*, 2009), In *Bacillis subtilis* Ca^{2+} , Fe^2 , Mg^{2+} enhanced the xylanase production (Annamalai *et al.*, 2009).

Effect of various carbon, nitrogen sources and metal ions, detergents and inhibitors on xylanase production are tabulated in Table 2.9

Table 2.9

Effect of carbon and nitrogen sources on xylanase production from various bacteria and fungi

Organism	Carbon Sources	Nitrogen Sources	Metal ions	Authors
	Stimulated	Stimulated		
<i>Aspergillus awamori</i>	Glucose			Botella <i>et al.</i> , 2005
<i>Aspergillus flavus</i>		Yeast extract Urea		Rukmani & Rajendran 2001
<i>Aspergillus foetidus</i>	Glycerol, Xylan trehalose			Shah & Madamwar 2005
<i>Aspergillus niger</i>	Starch	Ammonium sulphate		Haq <i>et al.</i> , 2002
<i>Arthrobacter</i>			Co ²⁺ , Zn ²⁺ , Fe ²⁺ , Cu ²⁺ , Mg ²⁺ , Ca ²⁺ , EDTA	Khandeparker and Bhosle 2006.
<i>Bacillus pumilus</i>		Olive oil, peptone, Yeast extract		Battan <i>et al.</i> , 2007
<i>Bacillus circulans</i>	Ribose			Qureshy <i>et al.</i> , 2002.
<i>Bacillus circulans</i>	Glucose, Fructose, Xylose			Dhillion <i>et al.</i> , 2000
<i>Bacillus subtilis</i>			Mn ²⁺	Yuan <i>et al.</i> , 2005
<i>Bacillus sp</i>	Glucose, Lactose, Fructose	Yeast Extract		Virupakshi <i>et al.</i> , 2005
<i>Bacillus sp.</i>			Ca ²⁺ , Mn ²⁺ , Fe ²⁺ , Hg ²⁺ , Mg ²⁺	Kaur <i>et al.</i> , 2015

<i>Bacillus sp.</i>		Yeast extract	EDTA	Csiszar <i>et al.</i> , 2001
<i>Staphylococcus</i> sp			Fe ²⁺ , Ni ²⁺ , Cu ²⁺ , Dithiothreitol	Gupta <i>et al.</i> , 2000

2.3.6 SCANNING ELECTRON MICROSCOPIC STUDIES (SEM)

The surface of untreated fibres appeared smooth, whereas that of the treated fibres was rough. These changes in pulp fibres resulted in its refining facilitating enhanced accessibility of chemicals used in subsequent bleaching stages. The fibre swelling also improved physical properties of the pulp. Similar distribution and separation of pulp fibres upon enzyme treatment by SEM was reported earlier (Poorna and Prema, 2007; Sanghi *et al.*, 2004).

2.3.7 BIOCHEMICAL PROPERTIES OF XYLANASE

MOLECULAR WEIGHT

Xylanases from different microbial strains and with the molecular weights in the range of 12kDa to 110 kDa are listed in the Table 2.10

Table 2.10

Characteristics of xylanases from different microorganisms

Fungi

Organisms	Optimum		Stability		Activity		Molecular Weight (kDa)	References
	pH	Temperature (°C)	pH	Temperature (°C)	pH	Temperature (°C)		
<i>Acrophialophora nainiana</i>	7	55	-	55			22.6, 22.1	Salles <i>et al.</i> , 2000
<i>Aspergillus niger</i>			5.0-6.0	60	5.5 & 6.0	45		Frederick <i>et al.</i> , 2004
<i>Aspergillus tamarii</i>	6.0-6.5	45						Gouda and Naby, 2002
<i>Aspergillus niger</i>	5	55					20	Li <i>et al.</i> , 2014
<i>Aspergillus</i>	6.5				6.0	55		Carmona <i>et al.</i> , 2007

<i>versicolor</i>								
<i>Aspergillus phoenics</i>		50-55						Rizzatti <i>et al.</i> , 2004
<i>Aspergillus carneus</i>	6	50			7.9		18.8	Fang <i>et al.</i> , 2008
<i>Aspergillus</i> sp S9	6-11	50-90			9	80		Sharma <i>et al.</i> , 2015
<i>Fusarium proliferatum</i>	5.0-5.5	55						Badal <i>et al.</i> , 2002
<i>Geotrichum candidum</i>	4	50	3-4.5	45			60-67	Rodionova <i>et al.</i> , 2000
<i>Trichoderma viride</i>						30		Nucbe <i>et al.</i> , 2012
<i>Shizophyllum commune</i>							26	Song <i>et al.</i> , 2013

BACTERIA								
Organisms	Optimum		Stability		Activity		Molecular Weight (kDa)	References
	pH	Temperature (°C)	pH	Temperature (°C)	pH	Temperature (°C)		
<i>Arthrobacter</i> sp					7-8	100	20	Khandeparker and Bhosle, 2006
<i>Bacillus coagulans</i>					4.5-10	45-75		Heck <i>et al.</i> , 2005
<i>Bacillus coagulans</i>					7			Chauhan <i>et al.</i> , 2006
<i>Bacillus licheniformis</i>							17,40	Damiano <i>et al.</i> , 2003
<i>Bacillus circulans</i>					6-8	50		Qureshy <i>et al.</i> , 2002
<i>Bacillus subtilis</i> CXJZ							23.3	Guo <i>et al.</i> , 2012
<i>Bacillus brevis</i>	7	55		45-90				Zheng <i>et al.</i> , 2014
<i>Bacillus halodurans</i>	9	70						Lin <i>et al.</i> , 2013
<i>Paenibacillus</i> sp	6	60					37	Zheng <i>et al.</i> , 2014
<i>Paenibacillus campinasensis</i>	7.5	60	5-9	70-80			41.3	Dheeran <i>et al.</i> , 2012
<i>Paenibacillus macerans</i>	4.5	50					205	Dheeran <i>et al.</i> , 2012
<i>Eschericia coli</i>		65-70	5-8	60				Yin <i>et al.</i> , 2007
<i>Cellulomonas flavigenia</i>		55-65			6.5			Herandez <i>et al.</i> , 2007
<i>Streptomyces cyaneus</i>					6	60-65	20.5	Ninawe <i>et al.</i> , 2008

2.3.8 APPLICATION OF XYLANASES

Xylanase (EC.3.2.1.8: endo- β ,1-4-D-xylanase) is mainly responsible for the hydrolysis of xylan with β -1-4-xylanolytic linkages. During the last decades xylanases have been received a great deal of attention mainly due to their various industrial applications such as pulp,papar,food and feed industries. Xylanase is responsible for hydrolysis of xylan, major hemicelluloses of plant cell wall(second most abundant). This enzyme is extensively used in food processing, chemical and pulp industries (Seyis and Aksoz,2005). The major uses of this enzyme are in biopulping,biobleaching,clarifying and liquefying fruit and vegetable juices. In paper and pulp industries, use of xylanase causes decrease in consumption of chlorine, absorbable organic halogen (AOX), Chemical Oxygen Demand (COD) and improves thereby the quality of wastewater.

2.3.8.1XYLANASES IN PAPER-PULP INDUSTRY:

Xylanases hydrolyse hemicellulose and they are of great interest to the paper and pulp industry because for their bleach boosting properties, which reduces chlorine consumption (Balakrishnan *et al.*, 1992; Viikari *et al.*, 1994). The following benefits of the usage of xylanases in the industrial sector have been demonstrated

- 1) Reduction of chlorine based chemicals and hydrogen peroxide.
- 2) Increase in the tear and burst strengths of the paper produced.
- 3) Reduction in the chemical oxygen demand in the effluent discharge lines (Beg *et al.*, 2000).

2.3.8.2 XYLANASES IN BREAD MAKING

Xylanases play an important role in bread making due to their water absorption capability and interaction with gluten. Utilization of xylanases improve bread qualities (good volume size,smooth texture, prolonged life time). In 2012, xylanases produced by *Bacillus licheniformis* were made commercially available by Novozyme as an additive for baking industry under the brand name “Panzea”. Microbial xylanases enhanced dough rheological properties as increase in loaf volume that improves its baking performance and so have great importance in cereal industry (Nunex *et al.*, 2001).

2.3.8.3 BIOBLEACHING AND BIO-SOFTENING OF FIBRES

Biobleaching and bio softening involves the treatment of fiber with microbes which have the ability to degrade the lignin content of the fiber. Lignin is the responsible for the dark colour, branching patterns and harshness. The spinnability of the fiber is greatly reduced because of poor elongation and high flexural rigidity. The treatment of lignocellulosic fiber with *Aspergillus flavus* for removal of lignin, pectin and other gummy material (Roy *et al.*, 2008). White rot fungi namely *Phanerocheate chrysosporium* and *Ceriporiopsis subvermispora* for removal of lignin (Jayapriya and Vigneshwaran, 2010) have been reported. For obtaining high quality textile materials with sufficient whiteness and hydrophilicity, it is necessary to remove pectin and lignin.

FTIR ANALYSIS

Various analytical methods have been used for studying of chemical and structural composition of fiber for example gas chromatography analysis of derivated sugars. However, a lot of time is spent at carrying out of such experiments, a plenty of sample is required. On contrary, FTIR is a rapid analysis that does not destroy interconnections of functional groups of cell wall components and obtain the information on structure in pictogram or even tomogram of the material (Stewart *et al.*, 1995). Now there is extensive information of analysis of chemical, structural and physical properties of various industrial crops fibers, including flax (Akin *et al.*, 1996).

FTIRbased peak intensity ratio is employed to differentiate the fiber types on the basis of the relative lignin and cellulose. In spite of a general similarity in the FTIR spectrum of lignocellulosic fibres, there are certain 'signatures' that can be assigned to specific components. The intensity of each can be considered as a representation of the proportion of components within the fiber.

The peaks at 3700-3400 represent the α -cellulose O-H groups and 2930-2900 represents the C-H stretching the cellulose. The peaks at 1740-1700 and 1250-1240 represents the C=O and C-O-C hemicelluloses stretching's. Two band ranges 1060-1050 and 1040-1030 denotes the C-OH, C-O of lignin. The band at 1690-1500 represents the oxygenous group of lignin (Vladimir *et al.*, 2010).

2.11 Applications of xylanase

Industry	Application	Function	References
Food Fruit and vegetable processing, brewing, wine production.	Fruit and vegetable juices, nectars and purees, oils (e.g., olive oil, corn oil) and wines	Improves maceration and juice clarification, reduces viscosity. Improves extraction yield and filtration, process performance and product quality.	Bhat, 2000
Baking	Dough and bakery products	Improves elasticity and strength of the dough, thereby allowing easier handling, larger loaf volumes and improved bread texture.	Maat <i>et al.</i> , 1992
Fruits	Fruit ripening	Softening of fruits during ripening, in ripening endoxylanases play an important role by modifying the polysaccharides in the cell wall matrix	Manenoi <i>et al.</i> , 2007
Feed Animal feeds	Monogastric (swine and poultry) and ruminant feeds	Decreases the content of non-starch polysaccharides (arabinoxylan), thereby reducing the intestinal viscosity and improving the utilization of proteins	Bhat, 2000; Nielsen <i>et al.</i> , 2008

		and starch. Improves animal performance, increases digestability and nutritive value of poorly degradable feeds, e.g., barley and wheat. Reducing emissions of N ₂ O and CH ₄ from manure	
Paper and pulp	Bio-bleaching of kraft pulps	Reduces chlorine consumption and toxic discharges and also reduces ClO ₂ it is contribution to global warming	Viikari, 1994; Beg <i>et al.</i> , 2001; Skales <i>et al.</i> , 2008
	Bio-mechanical pulping	Facilitates the pulping process and reduces the use of mechanical pulping methods, hence reduces energy consumption.	Bhat, 2000
	Bio-modification of fibers	Improves fibrillation and drainage properties of pulp, hence improving the process efficiency and the paper strength.	Bhat, 2000
	Bio-de-inking	Facilitates the de-inking process and reduces the use of alkali	Bhat, 2000; Frederix <i>et al.</i> , 2003
Starch	Starch-gluten separation	Reduces batter viscosity, improves gluten agglomeration	Frederix <i>et al.</i> , 2003

		and process efficiency	
Textiles	Biopolishing of fabrics, Retting of flax, jute, ramie, hemp, etc	Biopolishing-producing stonewashed look of denims. Enzymatic retting reduces/replaces chemical retting methods.	Beg <i>et al.</i> , 2001; Kalim <i>et al.</i> , 2015
Bioremediation/Bioconversion	Treatment of agricultural, municipal and food industry wastes	Treatment/recycling of wastes. Production of fermentable products, renewable fuel (bioethanol) and fine chemicals.	Prade, 1995; Saha, 2003
Pharmaceuticals	Xylooligosaccharides production	Reduction of cholesterol, maintenance of gastrointestinal health, improvement of the biological availability of calcium	Gupta <i>et al.</i> , 2014-15; Salupi <i>et al.</i> , 2015
Seeds	Germination of seeds	Germinating seeds naturally produce xylanases late in the germination process which might help to access required nutrients for the better growth of new plant	Kalim <i>et al.</i> , 2015

2.12 Commercial xylanases and their industrial suppliers

S.No	Suppliers	Product trade name	Application
1	Alko Rajamaki, Finland	Ecopulp	Pulp bleaching
2	Sandoz, Charlotte N.C and Basle, Switzerland	Cartazyme	Pulp bleaching
3	Clariant, UK	Cartazyme HS 10, Cartazyme HT, Cartazyme SR10, Cartazyme 9407E, Cartazyme NS 10, Cartazyme MP	Pulp bleaching
4	Genercor, Finland; Ciba Giegy, Switzerland	Irgazyme 40-4x/Albzyme 40-4x, Irgazyme-10A, Albzyme-10A, Multifect xylanase	Pulp bleaching, Baking food
5	Voest Alpine, Austria	VAI Xylanase	Pulp bleaching
6	Novo Nordisk, Denmark	Pulpzyme HA, Pulpzyme HB, Pulpzyme HC, Biofeed Beta, Biofeed plus, Ceremix	Pulp bleaching Feed Brewing
7	Biocon, India, Bangalore	Bleachzyme F	Pulp bleaching
8	Rohn Enzyme OY Primalco, Finland	Ecopulpx-100, Ecopulp x-200, Ecopulp x-200/Ecopulp TX-100, Ecopulp TX-200, Ecopulp XM	Pulp bleaching
9	Meito Sankyo, Nogaya Japan	Xylanase	Research
10	Rohm, Germany	Rholasc 7118	Food
11	Solvay Interrox, USA	Oplipulp L-8000	Pulp bleaching
12	Thomas swan, UK	Ecozyme	Pulp bleaching
13	Iogen, Canada	GS-35, HS-70	Pulp bleaching
14	Sankyo, Japan	Sanzyme Px, Alpelase F Sanzyme X	Feed Food
15	Enzyme development	Enzeko xylanase	Baking ,food,feed