Scientific Electronic Archives
Issue ID: Sci. Elec. Arch. Vol. 17 (3)
Mai/Jun 2024
DOI: http://dx.doi.org/10.36560/17320241910
Article link: https://sea.ufr.edu.br/SEA/article/view/1910

ISSN 2316-9281

Xylans as a promising prebiotic agent: a brief review

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Abstract. Xylans are the most abundant hemicelluloses in nature, constituting components of the secondary cell wall of plant cells.They are polysaccharides with versatile properties, which can be used in various industrial sectors.Several biological activities have been identified in different xylans, such as antioxidant, antitumor, antimicrobial, emulsifying, anticoagulant, and immunomodulatory activities. Among these applications of xylans, its use as a prebiotic stands out, composing functional foods. Xylans form non-digestible dietary fibers that serve as a source of energy for beneficial microorganisms, modifying the intestinal microbiome and bringing a general improvement to the health status of those who consume them. The literature reports the use of xylooligosaccharides as promoters of the growth of probiotic microorganisms, such as species of Lactobacillus and Bifidobacterium, in addition to treating dysbiosis and reducing intestinal inflammation. Due to their structural diversity, abundance in nature, and a wide range of biological properties, xylans are valuable components that can be used to create various products aimed at enhancing human health.

Keywords: Xylans, polysaccharides, prebiotics, functional foods.

Introduction

Hemicelluloses have aroused great interest in the scientific community because they are versatile polysaccharides, known for their bioactivity, biodegradation, biocompatibility, among other characteristics (Kapil et al., 2023). These properties suggest that these polysaccharides can be applied in various industrial sectors. They can be used as raw material for packaging, in the controlled release of drugs, as biodevices, and in the production of food films (Valladares-Diestra et al., 2023). Xylan is the main hemicellulose present in most plant species and can be extracted from various plants and different residues from the agricultural industry (Curry et al., 2023; Guo et al., 2023; Liu et al., 2023).

Structurally, all xylans have a basic structure of D-xylopyranose linked by β-(1→4) glycosidic bonds (Curry et al., 2023; Guo et al., 2023). This structure may undergo various substitutions depending on the origin and extraction method (Curry et al., 2023). Among these substitutions, α-D-glucuronic acid, 4-O-methyl-α-D-glucuronic acid and neutral sugar units such as α-L-arabinose, α-D-xylose and α-D-galactose (Guo et al., 2023). Based on the presence of these substitutions, xylans can be classified as homoxylans (only xylose residues) and heteroxylans (different linked monosaccharides) respectively (Liu et al., 2023).

The global market for xylan-derived products was valued at US$1.66 billion in 2022 and is expected to grow by 6% between 2023 and 2029, reaching around US$2.50 billion (Xylan Market, 2024). These data demonstrate the promising nature of the global market for products utilizing xylan as a raw material. Among the main products, its use as a prebiotic stands out.

Prebiotics are used as non-digestible food supplements because of their various health benefits, which include improvements in gastrointestinal, cardiovascular, neurological, inflammatory, oncological, and endocrine systems (Ferreira et al., 2023; Valladares-Diestra et al., 2023). These supplements induce a change in the composition of the intestinal bacterial population, promoting an increase in Lactobacillus and Bifidobacterium spp. The fermentation of these prebiotics by intestinal bacteria results in the production of short-chain fatty acids, such as acetate, propionate, butyrate, and lactate (Ferreira et al., 2023; Valladares-Diestra et al., 2023; Ravichandra et al., 2023).

The literature presents a variety of studies that explore the use of xylans as prebiotics. Authors such as Ravichandra et al. (2023), Cruz Filho et al. (2023), Yan et al. (2023), and Ismail et al. (2023) investigated the effects of xylans obtained from different sources, demonstrating their ability to induce the proliferation of various strains of probiotics. These studies offer valuable insights into the potential of xylans as prebiotics and emphasize their potential application in promoting gut health and overall well-being. Therefore, this review aims to present the prebiotic potential of different xylans.

Xylans

Hemicellulose is a component of the secondary cell wall of plants, constituting approximately 20% of the cell wall of plant species (Rennie & Scheller, 2014). This polysaccharide is divided into two main groups: xylans and mannans. The proportion of these macromolecules in plant species can vary according to the type and region of the plant from which it is extracted (Tryfona et al., 2023). Higher levels of xylans are generally found in eudicot plants, while mannans are more prevalent in bryophytes and pteridophytes (Smith et al., 2017; Tryfona et al., 2023). Due to the vast diversity of the eudicots group, consisting of approximately 200,000 plant species, this review will primarily focus on xylans.

Xylans are polysaccharides composed of a linear chain of β-1,4-linked xylose residues with a degree of polymerization ranging from 90 to 120 (Rennie & Scheller, 2014; Smith et al., 2017). These molecules have a double helical strand structure that binds to cellulose through hydrogen bonds, playing an essential role in the formation of the cell wall. This xylose residue can be replaced by glucuronic acid, methyl glucuronic acid, and arabinopyranose, depending on the function and location of these xylans (Smith et al., 2017). Therefore, they can be classified as monoxylans or heteroxylans depending on the presence of side chains.

These macromolecules also have a degree of acetylation that can vary from 20% to 60% depending on the species. The lack of these acetylations can weaken the cell wall and xylem vessels, as seen in Arabidopsis esk1, tbl32, and tbl33 mutants (Smith et al., 2017). Figure 1 illustrates the various structures of xylans.

In plant cells, xylans interact with other components of the secondary cell wall to provide rigidity and mechanical strength to the wall. Furthermore, xylans are fundamental for the absorption of water by the plant, both due to their hydrophilic properties and because they are one of the molecules responsible for xylem structuring. Xylans are also utilized by plants as a carbohydrate source for seeds. These properties have been observed in xylan-deficient species, which have shown structural fragility and xylem collapse (Rennie & Scheller, 2014; Ye et al., 2022).

The process of xylan synthesis in plant cells begins with the formation of carbon sources used for this process, which are UDP-GlcA, UDP-Xyl, and

UDP-Araf. UDP-GLcA is produced by UDP-GLc 6-dehydrogenase in the plant cytosol. Once formed, UDP-GLcA can be converted into UDP-Xly through UDP-Xly synthase. This enzyme is found in both the cell cytosol and the Golgi complex, with its cytosolic forms being the most significant for this conversion process. The production of UDP-Araf begins with the synthesis of UDP-Arap by UDP-Xyl 4-isomerase in the Golgi apparatus and UDP-Glc 4-isomerase in the cytosol and Golgi. UDP-Arap is then converted into UDP-Araf by UDP-aramutase. These molecules are sent to the Golgi apparatus by specific transporters (Ye et al., 2022).

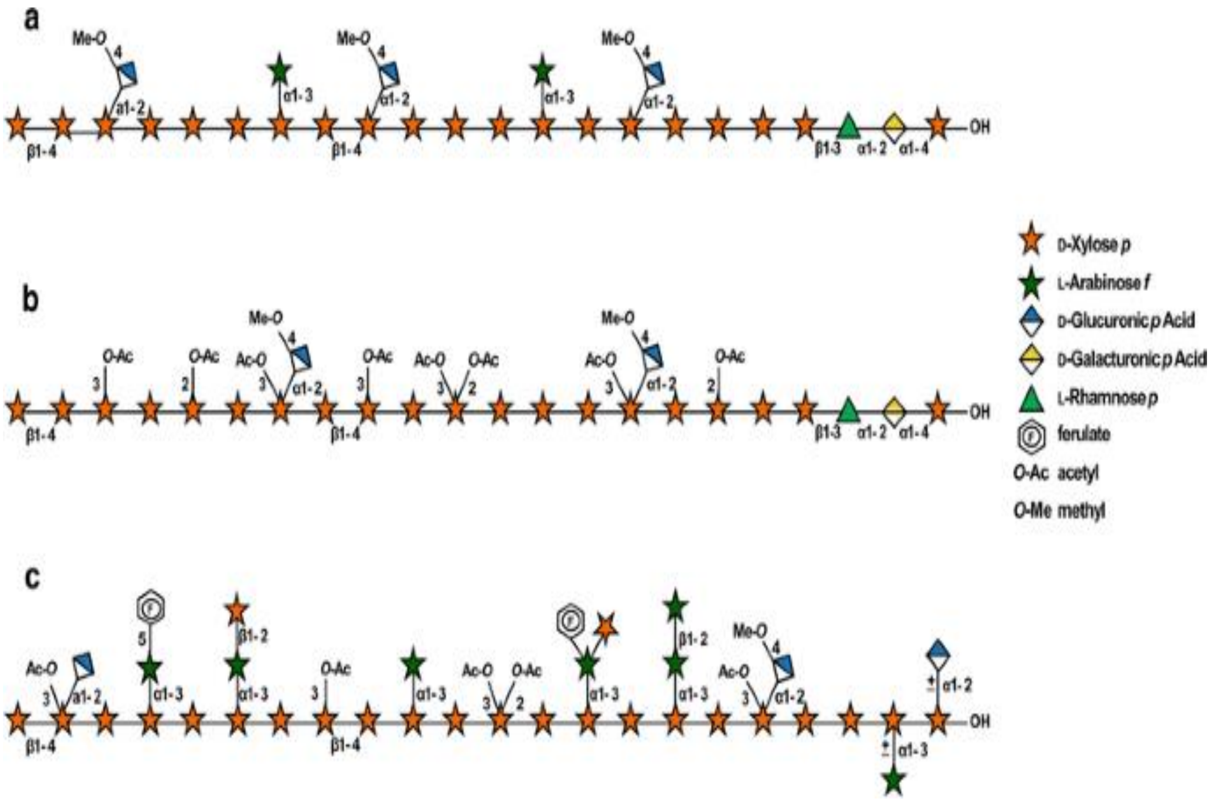

Figure 1. Xylan structures of secondary walls. Graphical representation of the main structural characteristics of (A) arabinoglucuronoxylan (B) acetylated glucuronoxylan (C) and acetylated glucuronoarabinoxylan (D) respectively. Font Smith et al., (2017)

Xylans are synthesized in the lumen and cytosol of the Golgi apparatus. Members of the glycosyltransferase 43 (GT43) family are proteins highly associated with the biosynthesis of the main chain, specifically IRX9 (irregular xylem 9) and IRX14 (irregular xylem 14), which are situated in the inner membrane of the Golgi apparatus (Rennie & Scheller, 2014). IRX14 and IRX9 are essential for the production of xylans, especially IRX9. When overexpressed, IRX9 was able to significantly increase the synthesis of xylans without relying on other GT43 enzymes. However, the mechanism by which these proteins act has not yet been elucidated.

The addition of side chains is carried out by xylanglucuronyl transferases (GUX), representatives of the GT8 family. This protein is responsible for adding glucuronic acid (GlcA) to the xylan side chain (Wierzbicki et al., 2019). After adding the side chain, this GLcA can be methylated with the help of glucuronoxylan methyl transferase, transforming GLcA into methyl glucuronic acid (Ye et al., 2022). Acetylations are primarily carried out by xylan O-acetyltransferase (XOAT), which is responsible for O-2 and O-3 monoacetylation, 2,3-di-O-acetylation, and 3-O-acetylation of 2-O-GLcA-substituted xylan residue (Wierzbicki et al., 2019). All these modifications that occur in the main chain are non-essential to the functioning of xylan molecules and may or may not occur in plant cells.

Biological Activities and Industrial Applications of Xylans

The biological activities of xylans are directly related to their structure. However, due to the complex and variable composition, the mechanisms of action are not fully elucidated when compared to other polysaccharides. The literature presents various biological activities induced by xylans (Figure 3). Cruz Filho et al. (2023) evaluated Amazonian xylans and identified antioxidant, anticoagulant, immunomodulatory, antitumor, prebiotic, and emulsifying properties. They also found that these xylans have low toxicity. Melo-

Silveira et al. (2011) found that xylans were capable of inhibiting the proliferation of tumor cells, as well as exhibiting antioxidant, anticoagulant, and antimicrobial activities *in vitro*.

Xylans have great industrial potential, being used as sweeteners, in the production of packaging, films, and fuels (Naidu&Hlangothi2018). In biomedical sciences, xylans can be used in the production of hydrogels, serving as a component for drug delivery and as an extracellular matrix in tissue engineering (Khaire et al., 2022). Hydrogels are primarily formed by polymeric mixtures, with the most common being a blend of xylan and acrylic derivatives. However, hydrogels containing chitosan are also observed. These copolymers will influence the ability of these hydrogels to retain water and their cytotoxicity. Changes in xylan molecules, such as the addition of aldehyde groups or the removal of arabinose, can enhance their interaction with other polymers, thereby improving the stability and permeability of these hydrogels.

Another important application for the food industry is the use of xylans as prebiotics (Singh et al., 2015). The human body is unable to degrade these polysaccharides because the human gene lacks the ability to decode xylanases (Valladares-Diestra et al., 2023). However, they are easily metabolized by the intestinal microbiota; therefore, xylans, along with many other carbohydrates of plant origin, pass through the digestive tract as dietary fibers (Singh et al., 2015; Valladares-Diestra et al., 2023). However, unlike other long-chain carbohydrates of plant origin, when in contact with certain microorganisms from the microbiota, this macromolecule can stimulate the production of cytokines, growth factors, and induce the secretion of other bioactive proteins (Cruz Filho et al., 2023).

Long-chain xylans are the most consumed and can promote the growth of Bifidobacterium pseudocatenulatum. Furthermore, the xylooligosaccharides released from the metabolism of these xylans mitigate intestinal dysbiosis by reducing the intestinal inflammatory response (Jain et al., 2015). Therefore, to ensure the prebiotic activity of these molecules, some tests must be carried out, with the main one being the evaluation of the growth of microorganisms. This evaluation will assess the degradation capacity of the isolated molecule by the microorganism, as well as its potential to interfere with bacterial growth (Singh et al., 2015; Valladares-Diestra et al., 2023). If it demonstrates negative interference, this molecule should not be used as a prebiotic. To date, no xylan or xylooligosaccharide subjected to this test has demonstrated any negative interference or hindered the growth of the bacteria it was tested on (Jain et al., 2015; Singh et al., 2015; Valladares-Diestra et al., 2023).

In addition to these, other tests such as evaluating degradation in simulated gastric juice and emulsifying properties are also important to determine the utility of these molecules (Cruz Filho et al., 2023). Degradation in simulated gastric juice aims, as the name suggests, to evaluate the degradation potential of the molecule when subjected to a pH similar to that of gastric juice in the intestines (Martins et al., 2023). The emulsifying potential will indicate an ability to facilitate the intestinal absorption of substances ingested together with xylan. It will also suggest a greater distribution of the molecule and, consequently, greater access of bacteria to them (Jain et al., 2015; Singh et al., 2015; Valladares-Diestra et al., 2023). In addition to these characteristics, xylans have demonstrated other properties that make them exceptional for the digestive tract.With anti-inflammatory properties and anti-tumorigenic capabilities (Sharma et al., 2020), xylans are an interesting option to improve the quality of life of patients with colon diseases, such as irritable bowel syndrome and colorectal cancer (Jain et al., 2015; Singh et al., 2015; Martins et al., 2023; Valladares-Diestra et al., 2023).

Application of xylans as prebiotics

According to a note published by the International Scientific Association of Prebiotics and Probiotics, prebiotics are molecules that enhance the performance of the intestinal microbiota, benefiting the host in turn (Ji et al., 2023). Xylans are considered prebiotics in the literature. They cannot be digested in the human body due to the lack of enzymes capable of metabolizing them; however, they are easily degraded by bacteria present in the gastrointestinal tract.

The utilization of these macromolecules as prebiotic agents has been explored in various studies over the past decade, focusing on xylans sourced from different plants, including those derived from wood industry byproducts. As shown in Table 1, they have demonstrated the ability to promote the growth of *Bifidobacteria*spp and *Lactobacillus*spp, which are part of the intestinal flora and are commonly used in probiotic formulations. These results indicate the need for studies to be carried out with other xylans to better understand the benefits of these molecules and determine whether structural differences will affect the results of biological activities.

Table 1: Utilization of Various Xylans as Prebiotics.

Source	**Results**	**References**
Xylooligosaccharidesfromsorghum (*Sorghum bicolor* (L.)	The xylooligosaccharides produced induced the growth of *Lactobacillus* T-10, indicating its prebiotic potential.	Ravichandra et al. (2023)
Xylans from the twigs and leaves of *Protium puncticulatum*	Regarding prebiotic activity in vitro, xylans were able to stimulate and promote the growth of different probiotics (*Lactobacillus casei* ATCC 25.180 and *Lactobacillus rhamnosus* ATCC 7469).	Cruz Filho et al. (2023)
Xylooligosaccharides from the mulberry branch	Xylooligosaccharides produced induced the proliferation of *Bifidobacterium adolescence* and *Lactobacillus acidophilus*.	Yan et al. (2023)
Encapsulated xylooligosaccharides	Encapsulated Xylooligosaccharides inhibit the proliferation of the probiotic *Lactobacillus acidophilus*	Ismail et al. (2023)
Sorghum xylooligosaccharides	These xylooligosaccharides acted as prebiotics, promoting the growth of the probiotic *L. acidophilus*	Nascimento et al. (2022)
Xylooligosaccharides obtained from beech wood xylan	These xylooligosaccharides promoted the proliferation of *B.adolescentisin vitro*	Ríos-Ríos et al. (2022)
Xylooligosaccharides obtained from industrial waste xylan	These xylooligosaccharides promoted the proliferation of *B. adolescentis*and *L. acidophilus in vitro*	Yan et al. (2022)
Xylooligosaccharides obtained from xylan extracted from rice husk and beech wood	These xylooligosaccharides stimulated the growth of organisms belonging to the genera Bacteroides, Megamonas and Limosilactobacillus	Gautério et al. (2022)
Xylooligosaccharides obtained from banana pseudostemxylan	These xylooligosaccharides stimulated the growth of the microorganisms *Lactobacillus plantarum* and *Lactobacillus fermentum*	Freitas et al. (2021)
Xylooligosaccharides obtained from corn cobs	These xylooligosaccharides stimulated the growth of the microorganisms *L. casei* TISTR1463, *L. delbrueckii* subsp. lactis TISTR1464 and *L. plantarum* TISTR1465	Boonchuay et al. (2021)
Xylooligosaccharides extracted from sugarcane residues (pith and peel)	These xylooligosaccharides stimulated the growth of the microorganisms Lactobacillus casei Shirota (LcS) and Bifidobacterium animalis subsp. Lactis ATCC® 700541™	Zidan et al. (2021)
Xylooligosaccharides Derived from *Eucalyptus nitens* Wood	These xylooligosaccharides stimulated the growth of *Bifidobacterium*spp	Míguez et al. (2021)
Xylooligosaccharides obtained from agricultural waste	These xylooligosaccharides stimulated the growth of three of the four probiotic cultures of *Lactobacillus* and *Bifidobacterium*	Ávila et al. (2020)
Xylan-type polysaccharides	Xylan-type polysaccharides were able to stimulate the growth of Bacteroides species and from *Bifidobacterium*	Zeybek et al. (2020)
Xylooligosaccharides obtained from sugarcane bagasse	These xylooligosaccharides stimulated the growth of *Lactobacillus acidophilus*, *Lactobacillus brevis*, *Lactobacillus viridescens*	Kaur et al. (2019)
Xylooligosaccharides obtained from wheat straw	These xylooligosaccharides stimulated the growth of *Lactobacillus brevis* DSM 1269	Faryar et al. (2015)
Xylobiose Obtained from Ragi Bran (Eleusine coracana, Indaf-15)	Xylobiose stimulated the growth of *Bifidobacterium* and *Lactobacillus*sp	Manisseri& Gudipati (2012)
Xylo-oligosaccharides obtained from Bengal gram (*Cicer arietinum* L.) bark and wheat bran (Triticum aestivum)	These xylooligosaccharides stimulated the growth of *Bifidobacterium adolescentis* NDRI	Madhukumar & Muralikrishna (2010)
Wheat arabinoxylan fractions	These arabinoxylan fractions stimulated the growth of different probiotic strains	Vardakou et al. (2008)

Conclusion

Xylans have a high market potential and can be used in various industries. They are utilized in the biomedical industry to create biofilms and hydrogels, as emulsifiers, sweeteners in the food industry, and for the production of biofuel in the energy sector. Furthermore, these macromolecules demonstrate antitumor, immunomodulatory, anticoagulant, antioxidant, emulsifying, and probiotic activities. This prebiotic activity has become increasingly relevant as these molecules help maintain gastrointestinal balance, which, in turn, influences the individual's overall health. However, these molecules can vary in the number of acetylations and functional groups, which can interfere with their biological and physicochemical properties. It is important to elucidate and understand the diversity of these molecules by evaluating their unique characteristics and capabilities.

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