



## Industrial Applications of Pectinases

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**ABSTRACT:** Application of pectinases in the commercial sector has been employed for nearly a century and due to the wide range of functions that these enzymes can work for, are making them critically important from industrial point of view. Pectinases are used in the industry on their role in the degradation of pectic substances aiding and enabling in overcoming the problems faced during the processing of purees, coffee and tea fermentation, fruit juices and in other food industry related manufacturing procedures. They break down the pectin content in the plants converting them to simpler molecules of galacturonic acid. The pectinases not only help in the food industry but also have remarkable applications in the textile, including retting, degumming, bio-scouring, maceration of plant tissues, paper making, and also has role in waste water treatment. Some of the roles of these pectinases solely and in conjunction with other enzymes e.g., amylases, xylanases, cellulases etc. have been comprehensively summarized in this review.

**Key Words:** Pectinase, degrading enzymes, industrial applications, xylanases, cellulases

### INTRODUCTION

The pectinases are delicate enzymes with three dimensional structures, (Fig 1) responsible for pectin hydrolysis into simpler sugar and galacturonic acid that is poly-galacturonic into mono-galacturonic acid. It is used for a row of enzymes, as a collective term, that is involved in the breaking of pectin (Kittur et al., 2003). According to Batten et al. (2007), out of overall manufacturing of enzymes 10% of production is occupied by the pectinases. Pectinases have been produced by many microorganisms (Sharma and Sathyanarayana, 2012; Sharma et al., 2013; Mohamadi et al., 2014). The enzymes are produced by microbes in combination with other enzymes (Kaur et al., 2011; Singh et al., 2015). The microorganisms are considered as primary source of industrial enzymes in which 50% being the fungi and yeast, 35% bacteria and 15% remaining have plant and animal origin (Anisa and Girish, 2014).

The plants have cell wall which provides an formidable barrier for any foreign invasion or infection. The cell wall has an integral part or

component called "Pectin" (Torres-Favela et al., 2003). It is composed primarily of galactouronans. The function of pectin is to cross link hemicelluloses and cellulose fibers, as a result of which rigidity of the cell wall is attained. It acts as a reinforce substance and is widely present in cereals, fruits and vegetables (Sathyanarayana and Panda, 2003). It is composed of modified sugars, galacturonic acid and carboxyl groups are esterified by methyl. Its major component is D-galacturonic acid (Aneja, 1996). Its highest concentration in the plant cell wall is present in the middle lamella and has a function of strengthening substance as described by Raju and Divakar. (2013). It is a complex polysaccharide having colloidal acid with backbones of galacturonic acid residue linked by  $\alpha$ -(1-4) linkages. The side chains consist of galactose, xylose, arabinose and L-rhamnose (Galiotou-Panayotou et al., 1993). Pectic substances on the basis of modification of the backbone chain are classified as: Protopectin, pectic acid, pectinic acid and pectin.

Degradation of the cell wall of a healthy plant due to invasion or infection by microbes could lead to devastating effects such as cell necrosis and tissue

maceration. The diseased and dead plants due to ecological changes are recycled back to their nutrient/ elemental level by microbes that have the ability to degrade the pectin component by producing globular proteins or enzymes (extracellular and intracellular) known as pectinases.

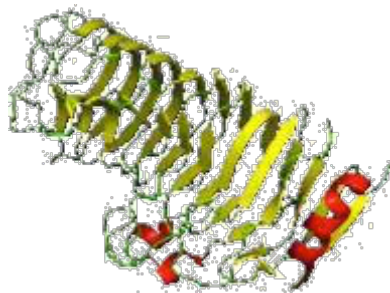


Fig 1: Computerized generated Image of polygalacturonases from *Aspergillus aculeatus* (1IA5) at pH 8.5 (Woo Cho et al., 2001).

### SUBSTRATE OF PECTINASE ENZYMES

The compounds on which the pectinolytic enzymes act are named using the generic name "Pectic substances". These substances are acidic, negatively charged polysaccharides found in the kingdom Plantae. They are a major component of middle lamellae and are found in between the cells in the form of calcium pectate and magnesium pectate.

Pectic substances are complex macromolecules linked glycosidically having high molecular mass and are found in higher plants. They are prominently seen in the middle lamellae (also seen in the primary cell wall), and are responsible for cohesion and structural integrity of tissues present in the plant body (Rombouts and Pilnik, 1980; Alkorta et al., 1998). Some of the pectic substances are soluble in warm water while the remaining might be solubilized upon boiling in dilute acids; oxalic acid in particular, or in alkaline EDTA, or ammonium oxalates (Zhbakov et al., 1976). Precipitation of pectin can be done by using ethanol from its aqueous solution to form a gel. Pectic substances are extractable from parenchymatous and meristematic tissues. These tissues comprise of 15-30% of cell wall material while the quantity is only 0.5-1.5% for heavily lignified tissues. The lignification begins in the primary cell wall and latter extends outwards to the lamella and inwards into the developing secondary cell wall (Zhbakov, 1964).

Their presence in the middle lamellae has been confirmed by the uptake of Iridium red by known Pectic substances, (Sterling, 1970) and also by the estimation of pectin by using alkaline hydroxylamine (Gee et al., 1959).

Table 1 shows the relative molecular mass of pectic substances ranging from 25 to 360 kDa (Sakai et al., 1993). Fogarty and Ward in 1972 reported the pectin content of some fresh fruits and some dried plant parts, a summary of which is given in the Table 2.

Pectic substances are complex colloidal acid polysaccharides having in its structure a backbone with side chain where galacturonic acid residues linked by  $\alpha$ -linkages constitute the backbone while side chains of pectin molecules consist of arabinose, galactose, L-rhamnose and xylose. The carboxyl groups of acid are partially esterified via methyl groups and can be partially or completely esterified by sodium, ammonium and potassium ions (Be Miller, 1986).

Pectic substances are labile which can hamper the studies of its primary structure. Extraction of these substances even in the mild conditions from the cell wall can result in artefacts that is fragments of complex compounds of polysaccharide and glycoprotein nature rather than in native molecules (Zhbakov, 1964). Dicotyledonous plants contain primary cell walls having 35% of pectic substances, but especially rich in this material are the (intercellular) middle plates.

Table 1: Molecular weights of some pectic substances (Sakai et al., 1993).

SOURCE	MOLECULAR WEIGHT (kDa)
Apple and lemon	200-360
Pear and prune	25-35
Orange	40-50
Sugar beet pulp	40-50

**Table 2: List of some fresh and dried fruits and plant parts having pectin content (%) (Fogarty and Ward, 1972)**

MATERIALS	PERCENTAGE OF PECTIN AS CALCIUM PECTATE	
	FRESH FRUITS	DRIED PLANT PARTS
Apples	0.5-1.6	-----
Apricots	0.7-1.3	-----
Bananas	0.7-1.2	-----
Citrus peels	-----	30-35
Currents	0.9-1.5	-----
Guavas	0.7-1.5	-----
Grapes	0.2-1.0	-----
Lemon peel	-----	35.5
Lemon pulp	-----	32.0
Lemon rind	-----	20.0
Pineapple	0.3-0.6	-----
Peas	0.5-0.8	-----
Peaches	0.3-1.2	-----
Potatoes	-----	2.5
Strawberries	0.6-0.7	-----
Sugar beet	-----	20-30
Tomatoes	0.2-0.5	-----

**PECTIC POLYSACCHARIDE GROUPS:**

There are several types of pectic substances (Glegg et al., 1974). These include:

1. Rhamnogalacturonans-1
2. Rhamnogalacturonan- 2
3. Homogalacturonans
4. Arabinans
5. Galactans
6. Arabinogalactans
7. Apigalacturonan

This clearly states that pectic substances have various forms in the plant tissues and this account as a probable reason for presence of various forms of pectinolytic enzymes (Jayani et al., 2005).

A committee that was appointed by the American chemical society in 1994 defined pectic substances as of the following 4 types: (Kilara, 1982; Alkorta et al., 1998).

- 1) Protopectin
- 2) Pectic acid
- 3) Pectinic acid
- 4) Pectins

**TYPES OF PECTIC SUBSTANCES**

1. Protopectin: These are water insoluble substances, from which soluble substances are produced, also referred to as parent pectic substance and yields pectin and Pectinic acid on restricted hydrolysis (Kilara, 1982).

2. Pectic acid: These are galacturonons containing negligible amounts of methoxyl groups. Normal and acidic salts of pectic acid are called pectates.

3. Pectinic acid: These are the galacturonons with variable amounts of methoxyl groups (>0 and 75%). Pectinates are normal or acid salts of Pectinic acid (Kilara, 1982).

4. Pectins: (Poly-methyl galacturonate): They are the polymeric materials, containing at least 75% of carboxyl groups of galacturonate units (Fig 2). Pectins are versatile, structural polysaccharides of plants. They are most prominently seen in primary cell wall and the middle lamella and occupies one-third of the dry weight of plant tissues (Gupta et al., 2008) Pectin as a plant component is present in non-woody parts of plant. Firmness and structure of plant tissues is acquired due to presence of pectin (Gummandi and Kumar, 2006). Pectin binds to the cellulose in the cell wall hence confer rigidity of the cell wall (Fig 3).

"Pectins" is the generic name used for the mixture of widely differing compositions containing Pectinic acid as a major component. In native form, it may be interlined along with other structural polysaccharides and proteins to form Protopectin (insoluble and located in the cell wall). It can be divided into 2 regions "smooth regions" and "hairy regions" (Fig 4). The source accounts for varying degree of esterification. It consists of 3 structurally well characterized motifs: HG, RG-1 and RG-2. They collectively form a network having potential for modulation in the structure as these degrading enzymes act on the cell wall.

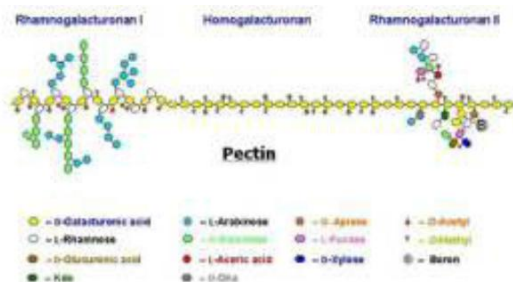


Fig 2: Structure of pectin (Harholt et al., 2010)

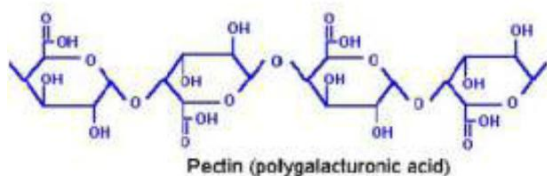


Fig 3: Structure of Pectin (Harholt et al., 2010)

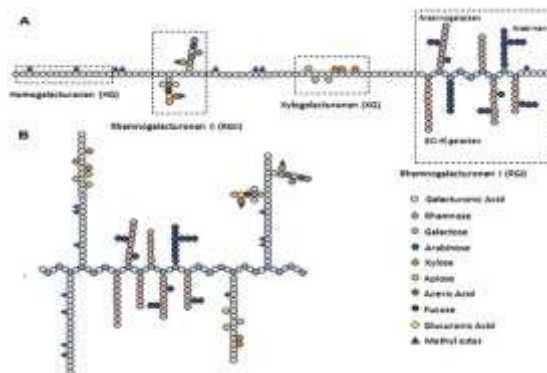


Fig 4: Schematic representatives of (A) conventional and (B) recently proposed alternative structure of pectin (Willats et al., 2006).

**Pectinase Production**

Pectinase as mentioned is produced and secreted by many plants and microorganisms. The list of organisms given below are examples in which PEs have been observed. (Table 3 & Table 4 given are compiled from article by Jayani et al. (2005).

**USES OF PECTINASES**

The functional applications of these enzymes in fruit juice and vegetable processing industries and other related industries has increased in the recent decade and more detail study on the pectinases and its wide applications has been reported by many research groups throughout the world highlighting them to be of importance

Table 3: Summary of a few reported microorganisms producing pectinolytic enzymes have been listed.

S.No	Microbial Species/ Strains	Reference in articles
1.	<i>Rhodotorula sp</i>	(Vaughan et al., 1969)
2.	<i>Saccharomyces cerevisiae</i>	(Galwors et al., 1994)
3.	<i>Phytophthora infestans</i>	(Forster, 1988)
4.	<i>Erwinia Chrysanthemi</i>	(Furuk and Heikkinen et al., 1992)
5.	<i>Aspergillus niger</i>	(Maldonado et al., 1994) (Maldonado and Saad 1998)
6.	<i>Pseudomonas Solanacearum</i>	(Schell et al., 1994)
7.	<i>Lachnospira pectinosticha</i>	(Cornica et al., 1994)
8.	<i>Penicillium frequentans</i>	(Kawano et al., 1999)
9.	<i>E. chrysanthemi 3604</i>	(Laurax et al., 2000)
10.	<i>Penicillium ischiaris</i>	(Hadj-Touh et al., 2002)
11.	<i>A. japonicus</i>	(Sentenova et al., 2003)
12.	<i>Lactobacillus sub specie coronis</i>	(Karam and Blarbi, 1995)

Table 4: List of plants that are able to produce pectinolytic enzymes.

S.No	Plants	References
1	<i>Vitis vinifera</i>	(Corredig et al., 2000)
2.	<i>Citrus sp</i>	(Arias and Burns, 2002)
3.	<i>Pouteria sapota</i>	(Arenas-Ocampo et al., 2003)
4.	<i>Carica papaya</i>	(T'ayyaz et al., 1993)
5.		<i>Lycopersicon esculentum</i> (Warlow et al., 1994)
6.	<i>Prunus mahis</i>	(Maldonado and Evans, 1996)
7.	<i>Malphigia glabra L.</i>	(Assis et al., 2004)

(Toushik et al., 2017). Following are some uses of pectinases in the commercial sector.

1. **Textile processing:**

The use of different kinds of chemicals was usually observed in many wet processes in the textile industry which caused pollution and also was corrosive to cause damage. Its use was of major concern but with the introduction of use of enzymes after isolation from several sources that can serve as an ecofriendly tool. Today, enzymes are considered as an integral part of the industry. Enzymes being employed in the textiles should have the following properties:

- a) Enzymes that accelerate the rate of reaction by lowering the activation energy and acts as a catalyst. i.e., remains intact at the end of the reaction.
- b) Enzymes that operate under mild condition i.e., should have an optimum temperature and pH at

which it works best, while at both ends of the optimum temperature it gets degraded.

- c) They should serve as an alternative to toxic, corrosive and polluting chemicals capable of eliminating the chance of causing any kind of carcinogenesis.
- d) They should have high degree of specificity for substrate and its activity should be easy to control.
- e) They should be biodegradable to allow proper prevention from pollution (Mojsov, 2012).

Pectinases can be used in the textiles as they can remove the cellulosic impurities from the fiber without causing any negative side effects on cellulose degradation (Hoodal et al., 2000). Applications of pectinases along with some other enzymes is used in fading of denim and non-denim, bio scouring , bio polishing , wool finishing peroxide removal, decolorization of dyestuff etc. Enzymes have been tried in every step of wet processing including in the treatment of effluent. Example of such enzymes can be given as in bio scouring or bio preparation using pectinases that target the non-cellulosic impurities within the fabrics (Lu, 2005). It is an eco -friendly tool for removing the non-cellulosic impurities from fiber with enzymes (Parveen and Suneetha, 2014).

## 2. Bio scouring of cotton fiber:

While fabrics are made from cotton the threads are coated with some adhesive substance that prevents the breaking of threads during the weaving. This is known as "sizing" and different kinds of compounds are used as a sizing agent. Starch due to being cheap was used as sizing agent. The removal of sizing agent is necessary before the fabric is processed further for dyeing. The de-sizing involves use of different chemicals such as alkali, acids and oxidizing agents. But their use can be destructing for the cotton as it not only removes the sizing agents but also degrades the fiber resulting in imperfection in dyeing and damage to soft feel of cotton. Pectinases are used in combination with other enzymes in the textile processing and has successfully been used to minimize the effects that were earlier seemed to hinder during the processing.

The pectinases along with amylases, cellulases, and hemicellulases when used removes the sizing agents in a safe and ecofriendly manner and has replaced caustic soda that was earlier used for the purpose. Studies on the bio scouring of cotton using acidic and neutral pectinases have been conducted (Pusic et al., 2015).

## 3. Clarification of juice and wines:

Pectinases contribute in removing the cloudiness from juices along with enhancement in the quality and flavour of the juice. There is an increasing in the preparation of such immobilized enzymes which can be employed in the clarification and depectinization of fresh juice to overcome the problems faced in the commercial processing (Cerreti et al., 2017). Pectinases are employed in industry for juices extracted from apple, orange, lemon, guava, grapes and many other fruits, use of pectinases in these fruits have been explained here briefly.

### a) Orange juice:

The pectin found in the orange juice gives it the particular appearance. Clarification of these substances is necessary to make them marketable and presentable on commercial level. The clearing of the pectic substances by degradation not only make the juice visibly clear but also aids in improvement of its yield overall. The classical methods that were been used involved heating or freezing, and were not good in respect to cost and quality of the juice that was extracted. Freezing method was expensive while heating spoiled the flavour of the juice (Braddock, 1981). The pectinases can be used in the orange juice industry to accomplish the following targets:

- 1) To soften peels so that peeling off of the citrus peels makes the processing of the juice easier (Ciechariska and Kazimierzak, 2006).
- 2) The pectinases are also applied in the crushing and clarification steps. The sediments suspended in the juice interact with the pectinase resulting in the juice being less viscous and increase yield due to the liquefaction that occurs during the

action of pectinases (Kareem and Adebowale, 2007).

They have been reported to increase volume to increase the volume of the juice (Kashyap et al., 2001). They have also been reported to impart critical role in the enhancement of the flavour of the processed juice.

#### b) **Lemon juice:**

Similarly, clarification step during lemon juice processing is aided with enzymes. Pectinases not only help in the extraction of juice but also benefit with increase yield of juice which is concentrated easily to be marketed, and reduction in processing time has also been reported (Prathyusha and Suneetha, 2011). The lemon juice was traditionally extracted relying on pectin esterase which was naturally present in the fruit content. The products obtained were majorly cloudy peel products. The peels with the pulp were ground into pieces of 3-5 mm and were heated after adding water in 1:1 ratio at a temperature of 95°C to destroy PME, but still the juice can have the pectic content, by using enzymes for degradation followed by centrifugation and pasteurization can process juice good enough to be concentrated and to be marketed.

#### c) **Apple juice:**

Apple juice can be obtained through a 2-step process i.e., firstly the treatment of the crushed apple with enzymes followed by pomace liquefaction treatment of with the pectinases and cellulases to completely extract the juice (Will et al., 2000). After washing and crushing, the apples are pressed to get the juice. The pectinases are added to facilitate juice extraction and pressing efficiency so that the mixture can be separated to be processed using sedimentation, filtration and centrifugation steps along the way to get the desired products (Fig 5). Process of production of apple juice has been shown in a comprehensive flow chart of the process. The juice is pasteurized to inactivate the enzymes in order to maintain the same cloudy texture of the fruit while centrifugation to acquire clear juice (Yamaski et al., 1964; Grassin and Fauquembergue, 1996; Kashyap et al., 2001). Polygalacturonases and pectin methyl esterase combinely are used for the proper clarification of the juice or pure pectin lyase

can be employed to get clarification (Ishii & Vokotsuka, 1973).

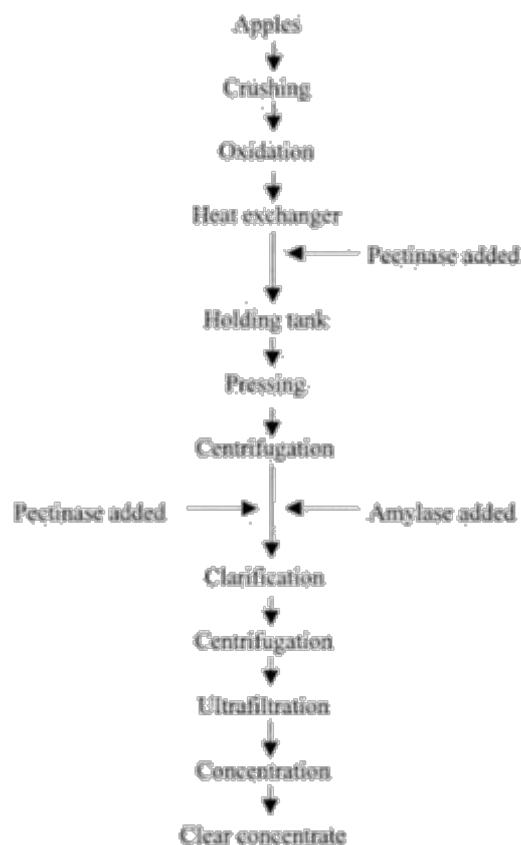


Fig 5: Flow chart showing the Production of apple juice (Grassin and Fauquembergue, 1996).

#### d) **Grape juice and wine making:**

Grape juice is either too sweet or too acidic in nature to be consumed in alone therefore it is consumed while mixing it with other fruit juices e.g., apple or in the form of mix fruit juices. Due to their high pectin content the grapes were earlier reported to be difficult to press and crush. The fruit after pressing and crushing was heated at 60-80°C in order to allow release of colour and the running juice released (with small solid particles in it) was subjected to filtration or centrifugation. Use of several enzymes like pectinases and hemicellulases can help in reducing the haze and gelling properties of pectin, along with removal of tartarate, to reduce the overall acidity of the grape juice to a considerate

level. The juice after treatment with enzymes can be filtered and then concentrated, pasteurized and bottled for use.

The treatment of macerated fruits with enzymes (before addition of the inoculum) improves the characteristics of the wine (Parveen and Suneetha, 2014). The pectinases can be applied to the grape juice processing or wine production in the following steps as summarized in the Table 5.

**Table 5: Use of enzyme in different steps of the juice/ wine processing summarized.**

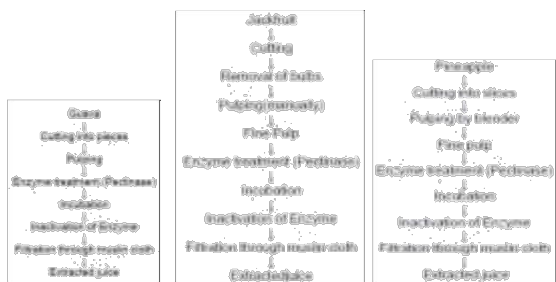
S.No	Step of juice/ wine processing	Enzymes used at level	Advantages	References
1	First step	During crushing	Reduced processing time, and increases volume of free juice released, juice is clarified and better release of anthocyanin.	(Preiel et al., 2000) (Pillnik and Voragen, 1970)
2	Second step	Before and after fermentation (in case of wine production)	Settles the suspended particles and removes undesirable microorganisms.	(Kashyap et al., 2001)
3	Third step	When wine is ready to be transferred and bottled.	Aid in clarity and increase filtration rate.	(Kashyap et al., 2001)

**e) Extraction of juice from jackfruit, pineapple and guava:**

Juices from jackfruit, guava and pineapple are turbid, viscous and grey in colour. Due to the change in the pattern of consumption of these fruits over the years in the population, has made them the alternatives to caffeine- containing beverages (Jagtiani et al., 1988; Sevda et al., 2012).

The commercial preparations of these juices require improvement in many ways as these juices can be preserved in the form of pulp/ purees. The single strength juice preservation is not economical so there is a need for alternatives to have bulk production of fruit juice without damaging the

flavour and texture of the juice in the processing. The fruit juices usually contain a water content ranging from 75%-90 % (Young, 1975). So, the fruit juice needs to be concentrated in order to attain microbial stability and for transportation of bulk juice by making it economical process as weight to volume ratio is reduced when juice is concentrated. Some issues related to the taste and colour were also reported as the juice extracted was viscous and had a sufficient amount of fiber content that contributed to its cloudiness as well. The problems faced in the classical methods of processing were observed to be resolved when enzymatic liquefaction was employed to these fruits using pectinases as the enzymes for treatment (Ahmed et al., 2014). It helped in the clarification and concentration of the juices. Pectinases not only helped in increasing the volume of the juice but also contributed in the solubilizing contents, color and aroma stability and clarifying the cloudiness by degrading the fiber content in these juices (Buchert et al., 2005). The clarified juices are more attractive and are generally acceptable to the population for use and can be processed for making of other fruit products like nectar, jelly and mix fruit juice etc. A summary of extraction of fruit juices from jackfruit (*Artocarpus heterophyllus*), pineapple (*Ananas comosus L.*) and guava (*Psidium guajana*) using pectinases have been summarized in the following flowcharts (Fig 6).



**Fig 6: Extraction of juices using pectinases in the enzymatic liquefaction process (Compiled from Ahmed et al., 2014).**

**f) Strawberry, raspberry and blackberry:**

The juices extracted from fruits like strawberry, raspberry and blackberry have high pectin which needs to be de-pectinized in order to get clear concentrates otherwise they will appear in the juice as residues (Will and Dietrich, 1992). Some extraction of juice from these soft fruits is possible

but difficult to process.

Sometimes the pectin & hemicellulose interact with other components such as proteins and form complexes that are not easily removed by the enzymes. These berries when treated with Rapidase BE in the maceration step helps in their juice extraction. Use of pectinase if necessary, ensures the removal of any residual left in the juice, followed by its filtration and stored at low temperature and dark conditions (Kashyap et al., 2000).

#### 4) **Preparation of purees:**

Pectinases can also be used in making purees from prunes, apricot, peaches, strawberries and fruits like mango, guava etc. the enzyme soften the skin and tissues (Tapre and Jain, 2014). The enzyme changes the fleshy pulp into semi-liquid product that is concentrated to 3 fold to form good textured puree to be sold as product. The puree produced in this way in case of guava can be used to make jams, syrups and juice blends etc. fruit pulp is treated with exogenous enzymes to yield juice. These enzymes have been reported to be used in the treatment of the pulp (Chaudhri and Suneetha, 2012; Khan et al., 2013).

#### 5) **Boosting aroma and flavour:**

Variety in aromas depends on the variety of grapes used and form the aromatic profile of wines. The aromas also originate with the yeast fermentation (Pineiro et al., 2006; Vilanova and Sieriro, 2006; Siero et al., 2012). The wines have aromatic components as well as the volatile components. The aromatic components contribute directly towards the scent of that wine (Williams et al., 1989; Bayanore, 1993; Winterhalter and Skouroumouvis, 1997). Use of pectinases help in breaking down the cell walls of grapes thus extracting their aromatic precursors. The interaction with these precursors via beta-glycosidase in the must, result in the increased components, enhancing the aroma of wines (Gomez-Plaza et al., 2000; Pinelo et al., 2006; Comitini et al., 2011; du Toit et al., 2011; Siero et al., 2012).

#### 6) **Maceration of plant tissues:**

The maceration process of plant tissues by

enzymes help in transforming the tissues into suspension with cells that are intact (Bock et al., 1983) and are used in the production of pulpy products such as food including juices, baby food, nectars etc. Enzymes such as pectinases or their use in combination with cellulases and hemicellulases is reported. The enzyme that specifically targets the middle lamella (Kashyap et al., 2001), the endogenous PE needs to be inactivated as it is very important for many macerated products (Dongowski and Bock, 1980). Enzymatic maceration results in limited degradation of the pectin components. This process can be used for carrots and dried instant potato mash (Boke et al., 1979; Bock et al., 1984).

#### 7) **Retting and degumming of plant fibers:**

Bast fibers are formed in the cortical region of plants in groups outside the pericycle, e.g., Ramie and sunn hemp. As they contain gum, it has to be removed before processing it for texture making (Hoondal et al, 2000). The bast fibers are required to be ret in order to be polished for its application in the textile sector e.g., in the production of commercial fibers. Retting is a process by which fibers in the form fiber bundles get separated from the cuticularized epidermis or woody core cells of the plant. The microbial activity in respect to the partial degradation contributes in the separation of cellulosic fibers from non-fiber tissues enabling us to extract the target resource for manufacturing, easily. Studies and work on retting of flax has clearly demonstrated the use and need of pectinases (Sharma and Van Sumere, 1992). Pectinases that are alkaline in nature are mostly used for the retting and degumming of jute, hemp, kenaff (Chesson, 1980; Bruhlmann et al., 1994). Retting is a process in which the pectin is decomposed by the bacteria and fungi while the fiber is released from the bark. The bacterial species from the genus *Clostridium* and *Bacillus* along with *Aspergillus* and *Penicillium* can be employed in the processing of the retting (Sharma and Robinson, 1983). Genus *Clostridium* species, *Clostridium butyricum* and *Clostridium felsineum* are regarded as major retting agents (Hellinger, 1953; Vonzyakovskaya et al., 1974; Kashyap et al., 2001). Pectinases have been used for retting of flax to separate the pectin from the fibers as reported by Hoondal et al. (2000). Retting of Latvian hemp sort "Purini" by use of pectinases has been reported by

Bernava. (2015). The treatment of pectinases along with xylanases is now days suggested to be economical as well as serves as the best alternative to the toxic and polluting chemicals (Kapoor et al., 2001). Study on bacterial pectinolytic enzymes, has been conducted that are used in retting and degumming of natural fibers (Chiliveri et al., 2016).

#### 8) Oil Extraction:

Oil extraction from citrus peels is done to obtain the citrus oils by using pectinases to remove the pectin content in these oil seeds which can cause emulsification hindering in the extraction process (Scott, 1978). Lemon oil is extracted via use of enzymes like pectinases. The extraction of these oils is done using an enzyme only or an extraction can be prepared by combination of more than 2 enzymes. Enzymes were used to degrade the cell wall usually in the grinding step or in the liquefaction procedure. For example, Table 6 shows a few enzymes used in combination to extract oil (Kashyap et al., 2001). Oil extraction from oil seeds like canola, coconut germ, seeds of sunflower, palm, olives and kernel were earlier done by using organic solvents. These solvents were not entirely beneficial as some were potentially injurious for health, e.g., hexane used in the process was a potential carcinogen, and therefore these had to be replaced by enzymes.

**Table: 6** Extraction of oil from oil-bearing material using enzymatic processing.

Oil seeds	Enzymes	Use level percent (w/w)	References
Rapeseed	Pectinase, Cellulase	0.1-3	(Deng et al., 1992)
Soybean	Protease	0.2	(Yoon et al., 1991)
Coconut	$\beta$ -Glucanases, Pectinase, $\alpha$ -amylase and protease	0.1	(Barrios et al., 1990)
Avocado	$\alpha$ -amylase	1.0	(Domiguez et al., 1995)
Sunflower	(Ultrazyme) cellulase, $\alpha$ -Galacturimidogalactanase	1.5	(Lanzani et al., 1975)
Peanut	Cellulase, protease	3.0	(Lanzani et al., 1975)

#### 9) Treatment of pectic wastewater:

The waste water usually contains pectin content when released out and this pectin is not degraded well by the microorganisms in the activated sludge treatment step. Treatment of such

waste water from citrus-processing industry was reported by using alkalophilic *Bacillus* sp (GIR 621) as reported by Tanabe et al. (1987). The treatment of wastewater with this enzyme has proved to be efficient in removing the pectic substances from waste water.

A soft rot pathogen, *Erwinia carotovora* (FERM P-7576) was reported to secrete an endo-pectate lyase which was reported to be effective in the pretreatment of wastewater but due to its phytopathogenicity, indirect pretreatment by enzyme produced from the bacteria was compared and reported, that all the pectin was solubilized that was present within the waste water (Tanabe et al., 1986). Similarly, the treatment of vegetable food processing waste contains pectin in the form of a by-product. Pretreatment with these enzymes facilitate the removal of pectin and makes it suitable for the degradation by activated sludge treatment (Hoondal et al., 2000; Jayani et al., 2005).

### CONCLUSION

Pectinolytic enzymes are produced by many organisms such as bacteria, fungi, yeasts and plants. Microbial pectinases are important in the decomposition of dead plant materials, contributing in the nutrient recycling e.g., in the carbon cycle. Microorganisms are major sources of enzymes. Microorganisms produce multiple pectinase forms which differ in their properties and molecular mass. Pectinases produced by bacteria and fungi are employed for commercial scale production of several food and textile related products. These enzymes are hydrolytic in nature and help in the degradation process majorly contributing to the food and juice processing industry. Pectinases are no doubt had their importance in past, have in present and will remain in future but there is always a need of faster, cheaper and heat stable enzymes in the industries. Therefore, more research is required to boost industrial applications of pectinases in an economical way.

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