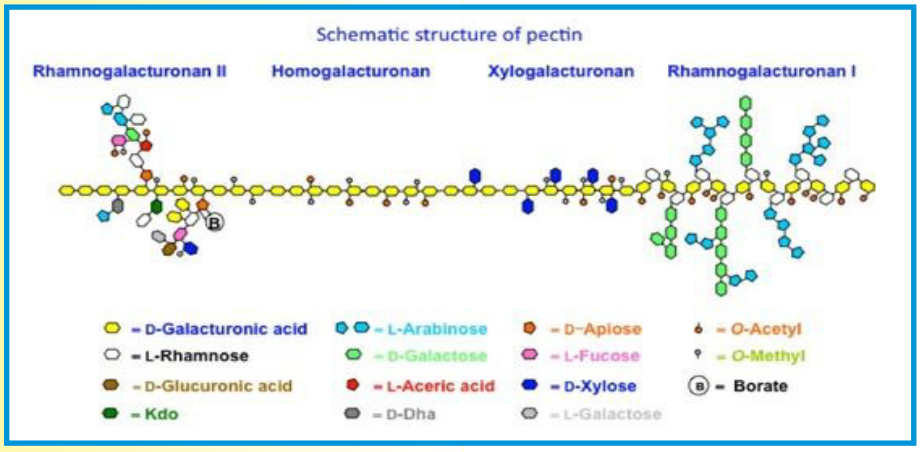
LIQUISEB and FIRMSEB for optimal pressing and firming fruit

- The choice of enzyme is a key for high quality fruit and vegetable processing
- Essential benefits for juice and vegetable producers
- A new generation of enzymatic preparations available
- LIQUISEB for high juice yield and high juice quality FIRMSEB PME for firming and softness of fruit and vegetables
- PECTINLYASE FOR FRUIT JUICE PROCESSING – LIQUISEB PRESS L 250

Pectin molecules are a group closely related to hetero-polysaccharides. The amount of pectin present depends on fruit variety and maturity. Insoluble pectin, with its gelling properties, increases the viscosity of fruit juice during pressing, thus slowing filtration and reducing yields.

Pectins are complex polysaccharides found in the cell wall of fruit. These polysaccharides include homogalacturonan, located in the so called *smooth zone* of the pectin molecule and the *hairy zone* is composed of rhamnogalacturonan I (RGI), and rhamnogalacturonan II (RGII). The hairy zone is highly branched containing arabans, galactans and arabinogalactans of various configurations and sizes are attached to the rhamnosyl units of RG. The Degree of Methylation ("DM") depends on the fruit variety and maturity. In apple and pear, the DM is between 65 to 92 %. The insoluble pectin, protopectin, is slowly transformed to soluble pectin by the action of endogenous pectinases present in fruit.

Apple and pear juice can contain anywhere from 2 to 5 g/L of pectins after pressing. During early season harvest, apple and pear juice contains starch and pectin. Pectinases and amylases must be added simultaneously after aroma recovery and pasteurization when the temperature reaches 45 to 60°C. These characteristics must be taken into consideration when choosing the correct enzyme balance for superior juice clarification, increased stability, prevention of browning and no production of methanol resulting from enzyme treatment.



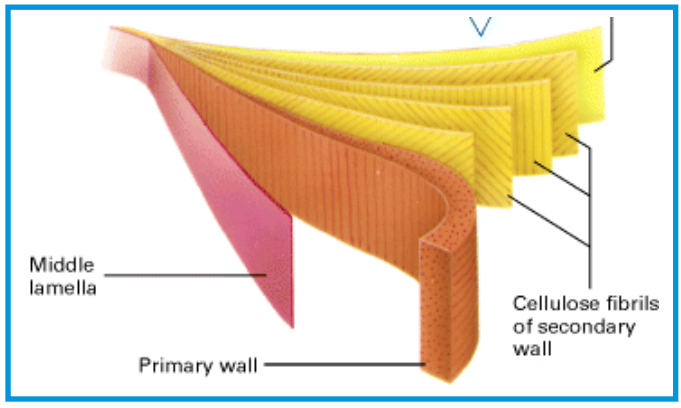
First step: Maceration and Extraction
Higher juice yield, dry pomace and no additional methanol

The cell wall of fruit will vary depending on fruit variety, climatic conditions and the duration of the storage of fruit. The cell wall possess three thin layers: middle lamella mainly composed of soluble pectin, primary wall "protopectin" and secondary wall.

Pectin is the major structural polysaccharide compound of fruit lamella and cell wall. This hydrocolloid possesses a great affinity for water and form gels under acid pH and sugar concentration. In the absence of the correct enzymes, insoluble pectin, with its gelling power, keeps juice in the pomace during pressing, reducing juice yield and pressability.

The apple cell wall is composed of pectin homogalacturonan and rhamnogalacturonan, hemicellulose (arabans, galactans, xylans and xyloglucans, among others), cellulose and proteins. Recent understanding of the different structural zones of pectins demonstrates the need for the presence of secondary activities to perform fragmentation in an efficient way (14-15). Indeed, different types of enzymes are necessary to degrade the homogalacturonan (smooth zone) and the rhamnogalacturonan (hairy zone). The exact composition of these zones largely depends on the type of apple and its maturity. Knowledge of fruit pectin structure is important for selection of the proper enzymatic preparation.

To optimize and respond to the needs of fruit and vegetable producers, Advanced Enzymes offers a new generation of enzymes for apple, pear juice processing and for fruit and vegetables firming. This new generation of enzymes are produced by the most advanced solid state fermentation techniques producing enzymes with robust activity.



LIQUISEB PRESS L250 was developed especially for apple mashing. This enzyme preparation contains high PectinLyase (PL) activity in addition to Pectin-esterase (PE), Polygalacturonase (PG) and Arabinase (AR). The

combination of these pectinase enzymes permits partial hydrolysis of the fruit while keeping a slight hardness to the cells for a better pressing and increased release of juice. The specificity of this enzyme preparation prevents excessive maceration, which in turn results in an improved pressing operation. Conveniently, **LIQUISEB PRESS L250** is optimal for various press systems used for apple juice extraction, including belt press, hydraulic horizontal press and others.

LIQUISEB PRESS L250 provides optimal mashing performance. Both juice yield and press capacity are significantly increased. In addition, the resulting juice quality is improved by the absence of galacturonic acid and cellobiose, which generally results from enzymatic

treatment and which can be responsible for juice browning. Essentially, the right combination of enzymes results in fewer colloids in juice, which leads to easier clarification and produces beautiful, golden apple juice.

The pomace left from the first pressing seems relatively dry. However, higher juice yield can be obtained by a second pressing. Increased extraction and separation of juice from solid can be obtained with the use of **LIQUISEB PRESS L250**. Not only will juice production increase, but brix will also increase. This is very important for juice that is concentrated. The total throughput is optimized preventing the juice from chemical oxidation and reducing the risk of microbial contamination.

Second step: Clarification and stabilization of juice

EXTRACTSEB RL is an enzymatic preparation produced by advanced solid state fermentation of *Aspergillus niger*. The enzymatic preparation is composed primarily of Endo-PectinLyase (also called PectinTransEliminase), acidic Endo-PolyGalacturonase and hémicellulases. The hemicellulases include endo-arabanases, arabinogalactanases and rhamnogalacturonase.

After maceration, juice can be clarified using **EXTRACTSEB RL**. At this stage it is important to avoid introducing secondary activities, particularly exo-enzymes, such as exo-Polygalacturonase, cellobiase and others. These "exo" enzymes may reduce the quality of the finished juice. The enzyme composition of **EXTRACTSEB RL** provides optimal clarification through complete degradation of the remaining soluble and other neutral cell wall colloids before filtration and ultrafiltration. At the earlier stage of fruit maturation during the beginning of the season, unripe apples may contain up to 15% starch. The quantity of starch decreases during processing, but around 5 per cent usually remains. Starch creates an undesirable haziness in both single strength and concentrated apple juice. Fortunately, starch can be hydrolysed during clarification by using an adapted dosage of acid -amylase enzyme, like **SEBAMYL GL400**. This enzymatic activity is active at the lower pH consistent with that of different apple varieties.

For extraction, clarification and stabilization of apple juice concentrate, **LIQUISEB PRESS L250**, **EXTRACTSEB RL** and **SEBAMYL GL400** all work in synergy by providing a solution for each step of apple and pear juice processing. **EXTRACTSEB RL** and **LIQUISEB PRESS L250** are very robust pectinase blends due to the microorganism selected and the solid state fermentation system used for its production. The efficiency of **LIQUISEB PRESS L250** during mashing is due to the high activity of PectinLyase, The addition of arabinase and arabinogalactanase in **EXTRACTSEB RL** ensures fast action with a lower dosage. **SEBAMYL GL400** eliminates starch haze.

LiquiSEB Press L 250 and **EXTRACTSEB RL** ensure high degradation of oligomers present in apple juices, which helps speed filtration and ultrafiltration processes. Still, during processing, pectin/protein complexes can be released from the cell wall. This pectin/protein complex causes significant filtration and UltraFiltration membrane fouling, which in turn decreases filtration efficiency, even after cleaning with traditional methods. Pectins and other neutral polysaccharides combined with proteins form a thin film on filter membranes that are difficult to remove. **SEBCLEAR UFL** is formulated to prevent the accumulation of colloids plugging the UF membrane pores.

SEBCLEAR UFL acts in synergy with ExtractSEB RL given their complementary activities, which includes acid protease, arabinase, arabinogalactanase and rhamnogalacturonase. The presence of these activities permits stabilization of concentrated apple juice by eliminating arabinogalactanproteins (AGP). The essentials of clarity and stability for apple juice concentrates are essential, especially in the case of exportation.

BENEFITS:

FIRMSEB is a highly purified pectin methylesterase produced from selected strains of Aspergillus niger (non-Genetically Modified Micoorganism).

FIRMSEB is produced by using a solid state fermentation system (SSF mode).

The PME activity generated by using selected strains and SSF system gives an extensive pectin de-esterification and increased calcium cross-linking of the pectins in the middle lamella.

FIRMSEB improves texture, viscosity, mouthfeel and taste of many different types of fruit such as strawberry, cherry, apricot and tropical fruit.

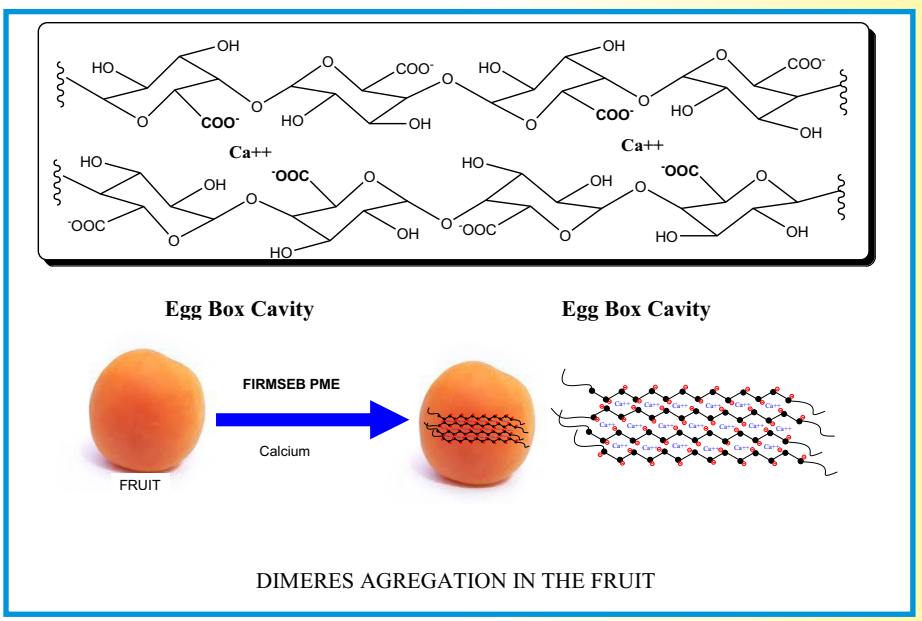
FIRMSEB improves firmness and taste of processed tomato, onion and other fruit and vegetables.

PectinMethylEsterase for fruit firming - FIRMSEB PME

FIRMSEB PME is a new generation of pure PectinMethylEsterase (PME). It is produced by a non-genetically modified (nonGMO) microorganism using a solid state fermentation (SSF) system. **FIRMSEB PME** is free of other pectin depolymerases such as endo-pectinlyase, endo- and exo-polygalacturonase or rhamnogalacturonase.

The food processing industry produces whole fruit and fruit pieces in various preparations. Food processors can either work with noble fruit or with chunks of frozen fruit from different origins. The advantage of the second solution is cost, which obviously is less for fruit chunks. However, in this type of preparation, pieces are damaged, lose water or liquefy completely during the process (thawing, heating, agitation, pasteurization). Processing and cooking fruit and vegetables can significantly impact their structural integrity in a negative way, whether whole, in slices, pieces or purees. Cooking can change texture resulting in a loss of firmness, shape, color and other attributes that make the end product less desirable. **FirmSEB PME** de-esterifies pectin molecules, increasing reactive carboxylic acid groups in the pectin molecular chain. This process then allows the formation of calcium bridges between pectin molecules, which strengthens the pectin molecular network and, in turn, the structural integrity of the fruits and vegetables being processed. **FirmSEB PME** is specifically designed to enhance the structural integrity of processed vegetables and fruit, whether whole, in pieces or purees. This includes canned vegetable, sauces, tomato sauce, tomato pieces, fruit for baking, jams, preserves, sauces, yogurt and many more.

The illustration below demonstrates how FIRMSEB PME enzyme acts with calcium to create strong calcium pectate complexes. The firming effect involves free carboxyl groups binding with calcium divalent cations between pectin polymers. A cross-linking with pectic polymers may limit the access of hydrolytic enzymes to wall components, particularly hemicelluloses, which in turn avoids liberation of pectin that is linked covalently to hemicelluloses. As a result, the linkage of the middle lamella with the primary wall is maintained. Thus, the cell is maintained as well as cohesion and fruit firmness. Further, the presence of calcium in fruit inhibits the activity of exo-PG and endo-PG. The calcium pectate created by PME eliminates the negative effects of mechanical and thermal effects during fruit processing. Many vegetables and fruits can be processed by using FIRMSEB such as tomato and other vegetables, peach, mango, strawberry, raspberry, apple, pear.



DIMERES AGREGATION IN THE FRUIT

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