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Effect of different extraction conditions on yield of pectin extracted from *Prunus armeniaca*

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Abstract
Pectins have been utilised for their functionality in foods for many years. Pectin is a complex mixture of polysaccharides that makes up about one third of the cell wall dry substance of higher plants. Much smaller proportions of these substances are found in the cell walls of grasses. The highest concentrations of pectin are found in the middle lamella of cell wall, with a gradual decrease as one passes through the primary wall toward the plasma membrane. Pectin has a long history as a food additive. The current study was done to optimize the pectin extraction during various stages of ripening in *Prunus armeniaca.* The optimization of pectin extraction was done by varying time (30 min, 60 min, 120 min), pH 2, 3, 4, 5) and ER (Ethanol ratio 01:01, 01:02, 01:03). The highest yield of pectin was observed at pH 2 and ER (01:01) at the incubation time interval of 120 minutes. However a decrease in the yield of pectin was observed during ripening. Maximum yield obtained at pH 2 and ER (01:01), and at incubation time of 120 min was in Immature green stage of fruits (0.78%), then the yield decreased consecutively in Mature green(0.64%), Pre ripe (0.58%) fully ripe(0.35%).
Keywords: Pectin, Ethanol Ratio.

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INTRODUCTION

Prunus armeniaca L. is a tree classified under *Rosaceae* family [2] and is mostly cultivated in Korea, China, India, Japan, Iran, North Africa and United States of America [3]. Pectin is a complex mixture of polysaccharides that makes up about one third of the cell wall dry substance of higher plants. Much smaller proportions of these substances are found in the cell walls of grasses. Pectin has a long history as a food additive. Fruit ripening is a very complex process. A large number of biochemical, physiological, and structural changes occur during the ripening of fruits. Some of the biochemical changes are the synthesis and actions of those hormones responisble for ripening, the degradation of chlorophyll, the biosynthesis of carotenoids, anthocyanins, essential oils, and the metabolisms of those sugars, acids and volatile compounds involved during taste and flavour development [4]. In the early developmental stage, fruit tissues undertake several rounds of cell division, followed by nutrient, metabolite, and energy storing during cell expansion[5,6] .The frutis are subsequently ripening and undergoing a series of changes that starch converts into small molecules, such as easily absorbed compounds: monosaccharide and volatile secondary metabolites[7]. *Prunus armeniaca* is a very tasty fruit consumed by people, it has been used for preparing jams and jellies through years but cell wall polysaccharides have not been studied in this during ripening. So this plant was selected for this study. The plant was identified from Botanical Survey of India, Dehradun. The fruits of *Prunus armeniaca* were collected during four stages of ripening viz- Immature green, mature green, pre-ripe and ripe stages. Further study was conducted to optimize the pectin extraction.

MATERIALS AND METHODS

Extraction of Pectin

Pectin was extracted by a slight modification in the method of Tyang *et al*., 2011. [8] Extraction of pectin from the fruits during various stages of ripening was done sequentially as follows-

a)Blending

About 10 g of fruit pulp of *Prunus armeniaca* Linn.was taken from all stages of ripening viz. immature green, mature green, pre – ripe and ripe stage was taken and blended with 250 ml distilled water.

b) Variation of pH

The extraction was performed under different pH environments ranging from 2-5. In order to get the desired pH, the mixture of all the stages of *Prunus armeniaca.* was then acidified by adding 40% citric acid. The mixture was stirred continuously and pH was monitored with the help of pH meter. Continuous stirring of the mixture was done to get the desired pH When appropriate pH was maintained the beaker containing acidified mixture of blended Fruit pulp of all the stages of *Prunus armeniaca* were transferred to water bath maintained at 60^0C.

c) Variation of time

By keeping the volume of the mixture constant the extraction of pectin from all the stages of *Prunus armeniaca* was carried out in a time-dependent manner at three different incubation times, 30, 60, 120 min in the water bath. After incubation, the resulting mixture was first filtered with cheesecloth and then with Whatman No. 1 filter paper. The final extract was then evaporated to one-fifth of its initial volume by using rotary evaporator[9]. The concentrated extract was subsequently filtered by a Whatman no. 1 filter paper to remove any excess insoluble impurities completely.

d)Precipitation of pectin

Precipitation was pectin was done by 95% ethanol. Ethanol was added to the mixture and the mixture was kept for 24 hrs at room temperature for pectin floatation and to facilitate the equilibrium between colloid –liquid phase. The ratio of *Prunus armeniaca*. Fruit pulp to ethanol volume (ER) was 01:01, 01:02, and 01:03. For complete separation of the precipitated material the solution was centrifuged for 15 min at 9000 rpm. The supernatant was discarded and the residue deposited at the bottom was washed with 45% ethanol to remove impurities like monosaccharide and disaccharide. Then solution was centrifuged at 9000 rpm for 15 min to separate pectin substances from the liquid medium.To achieve exact and pure amount of pectin, it was washed and centrifuged a few more times. The pectin extracted was freeze dried and stored at - 20°C until required.

Calculation of yield of pectin

The dried pectin extracted under different extraction conditions was weighed and yield was calculated. The % yield was expressed as mass of dried extracted pectin /mass of dried fruit pulp.The % yield was calculated and expressed per 1g of dried fruit pulp.

RESULTS

Yield (%) of pectin extracted at pH 2 at different time intervals by varying ethanol ratio during ripening in *Prunus armeniaca*.

At pH 2 the % yield of pectin showed promising results. The maximum yield was obtained at ER 01:01 at the time interval of 120 minutes. However the yield of pectin was decreased during ripening. The yield was maximum in Immature green (0.76%), then the yield decreased consecutively in Mature green(0.64%),Pre ripe (0.58%) fully ripe(0.35%). The same is depicted in Fig No.1.

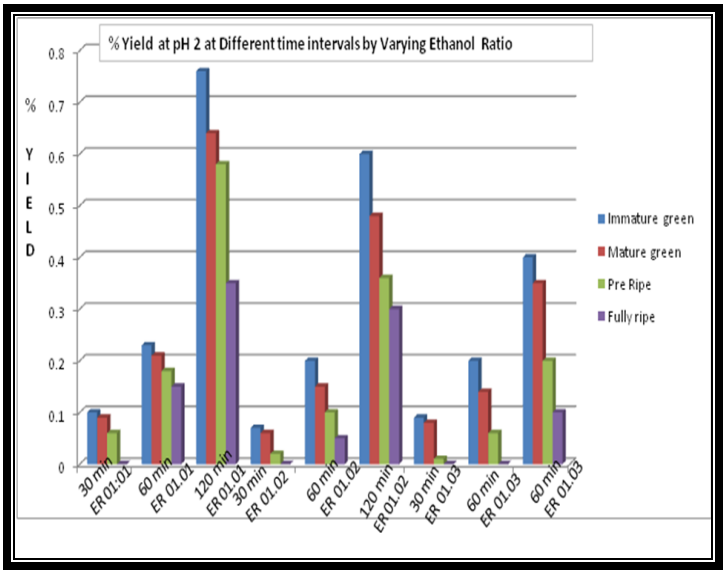

Fig: 1 Yield of pectin at pH 2 during ripening in *Prunus armeniaca* by varying extraction time and Ethanol Ratio.

Yield (%) of pectin extracted at pH 3 at different time intervals by varying ethanol ratio during ripening in *Prunus armeniaca*.

At ph 3 yield of pectin showed less promising results as compared to the yield obtained at pH 2. However maximum yield at pH 3 was obtained at the time interval of 120 min and ER 01:01. The yield was decreased during ripening. Immature green showed maximum yield (0.55%), Mature green showed a yield less than immature green (0.34%). The yield was consecutively decreased in pre ripe (0.2%), fully ripe (0.05%). The same is depicted in Fig. No.2

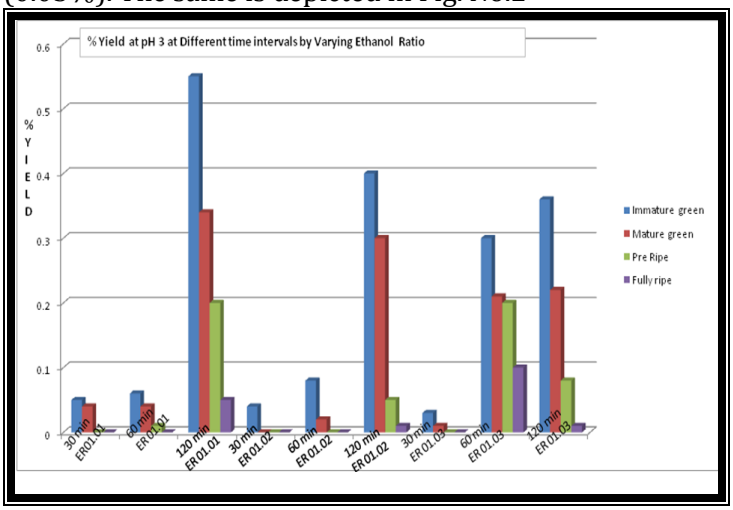

Fig:2 Yield of pectin at pH 3 during ripening in *Prunus armeniaca*, by varying extraction time and Ethanol Ratio

Yield (%) of pectin extracted at pH 4 at different time intervals by varying ethanol ratio during ripening in *Prunus armeniaca.*

Though the maximum yield of pectin at pH 4 was observed at ER 01:01 and time interval of 120 min, but it does not showed promising results. The yield was decreased during ripening Immature green(0.5%), Mature green (0.3) , Pre ripe(0.15), Ripe(0.09) but it was very low as compared to the yield obtained at pH 2.The same is depicted in Fig No 3

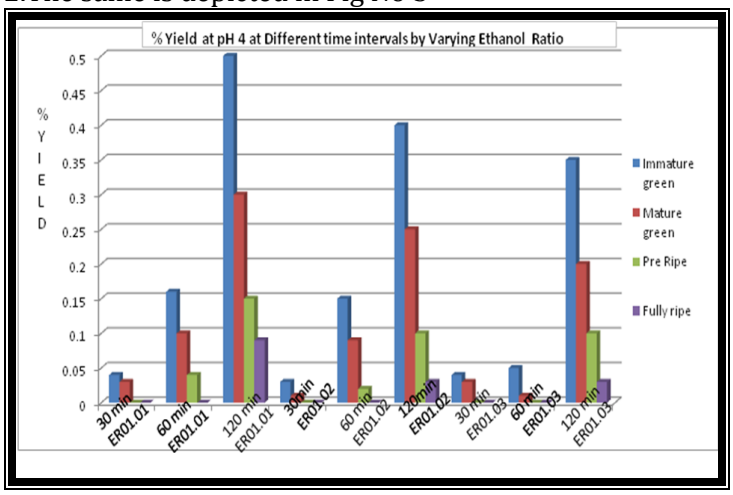

Fig:3 Yield of pectin at pH 4 during ripening in *Prunus armeniaca*, by varying extraction time and Ethanol Ratio

Yield (%) of pectin extracted at pH 5 at different time intervals by varying ethanol ratio during ripening in *Prunus armeniaca*.

The yield of pectin obtained at pH 5 was too low, the yield showed the same decreasing pattern during ripening, viz. immature green (0.4%), Mature green (0.2%), pre ripe (0.12%), Ripe (0.03%). Time and ER showed the same pattern for pH 5, Maximum yield was obtained at time interval of 120 min at ER 01:01. The same is depicted in Fig No.4

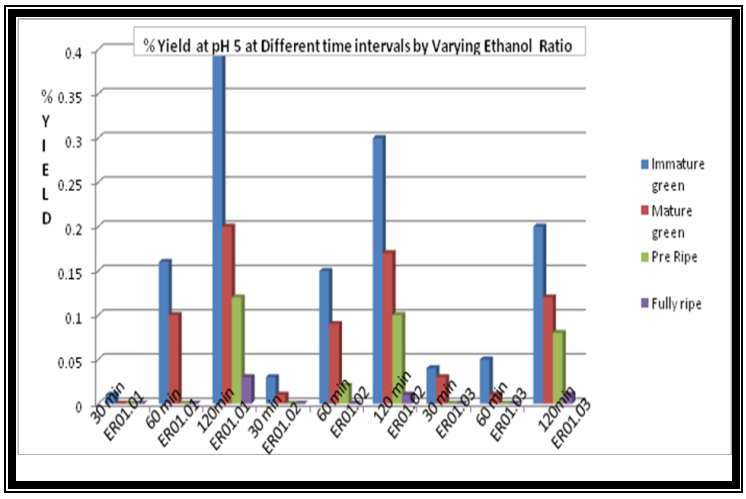

Fig:4 Yield of pectin at pH 5 during ripening in *Prunus armeniaca*, by varying extraction time and Ethanol Ratio.

DISCUSSION

pH is an important factor to be controlled in pectin extraction[10].This study indicated that pH 2 is best for extraction of pectin from *Prunus armeniaca*. Though *Prunus armeniaca* has not been studied yet in such a manner but these findings can be correlated with the extraction of pectin from other fruits as stated by Masmoudi et al., 2008 [11] (optimum pH for pectin

extraction falls between pH 2 to 2.8). However, the optimum pH for the extraction of pectin from *Prunus armeniaca* in present study contradicts the results of Wang et al, (2007)[12], and Yujaroen et al., (2008)[13], which reported that optimum pH for extraction of pectin is 1.01 and respectively. Also Pectin precipitation is governed by the addition of suitable amount of alcohol (ER) into pectin solution, which can increase the yield[13]. The maximum yield in this study was observed at ER 1:1. Maximum yield of pectin was observed at the time interval of 120 min.

CONCLUSION

From the current study it was concluded that pectin yield decreases during ripening in *Prunus armeniaca* and the best yield is observed at pH 2, ER 01:01 at the time interval of 120 min.

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