Browning in Sago Logs (*Metroxylon sagu*): Mechanism and its Prevention

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Introduction

Sago palm is considered as a main source of starch in South East Asia. In Malaysia, sago palm plantations and sago starch industries are located in Peninsular Malaysia and Sarawak covering a total area of 20300 ha with a productivity of 10-12 tons/ha of sago starch annually (Department of Statistics et al., Malaysia, 1997). It is reported that the pith contains almost 88 % starch (Encyclopaedia Britannica et al., 1997). There are two main species cultivated in Malaysia; the *Metroxylon sagu* and *Metroxylon rumphii*. The starch, despite having commendable properties and relatively least expensive in the market, it suffers some losses due to browning which deteriorates its quality. Though this could be attributed to the poor processing conditions in the sago factories, a substantial amount of change in the colour of starch is believed to take place during the storage period. The objectives of the project were therefore to identify causal factors of browning in sago logs and sago starch and also to develop the effective preventive measures during transportation, processing and storage of the logs/ starch.

Materials and Methods

Sago (*Metroxylon sagu*) pith was collected from Batu Pahat, Johor. The tree used in this study was between 10 to 11 years old. Analysis and identification of phenolic compounds were done by HPLC. Protocols for purification of polyphenol oxidases and peroxidases were developed at the Department of Food Science, UPM, Serdang, using a combination of cation- and anion-exchange chromatography and gel filtration chromatography. Enzyme activity was assayed using spectrophotometer and oxygen electrode

Results and Discussion

Factors that contributed to the browning in sago (*Metroxylon sagu*) were studied. Two enzymes responsible for the browning reactions, membrane-bound polyphenol oxidases (LPPO1 and LPPOII) and membrane-bound peroxidases (LPOD1 and LPODII), were successfully isolated and purified to homogeneity from fresh sago pith by a combination of osmotic shock, Temperature Induced-phase Partitioning and cation-and anion-exchange chromatography and gel filtration chromatography. LPPO1 and LPPOII had a molecular weight of 39.5 and 40.0 kD, respectively. Both isozymes showed variable sensitivity to heat treatment. The LPPO1 and LPPOII were also highly sensitive to treatment with detergent (SDS), alcohol (ethanol) and protease (trypsin). Their activities were several folds enhanced by the treatments. Identification of the phenolic compounds in sago logs using HPLC revealed the presence of four major phenolics namely catechin, catechol, epicatechin and 4-methylcatechol. The LPPOs were highly reactive towards the diphenols in the following order: 4-methylcatechol, epicatechin, catechol and chlorogenic acid. The enzymatic reactions were found to be more favorable in the regions between pH 4.5 and 5.0.

As for peroxidases (POD), two isozymes namely LPOD1 and LPODII with molecular weights of 33.0 and 46.3 kD, respectively were purified to homogeneity. The enzymes showed high activity at pH 5.5 – 6.0 with guaiacol as a substrate and were highly reactive towards tetramethylbenzinidine, chlorogenic acid, guaiacol and pyrogallol in the presence of H_2O_2 but not reactive towards catechol and 4-methylcatechol. *p*-coumaric acid at a concentration of 20 mM was found capable of inhibiting 100 % activity of both LPPO and LPOD while L-cystein, kojic acid and ascorbic acid were less effective in inhibiting the enzymes. Tree maturity had a significant effect on the concentration of phenolic compounds and the LPPO enzymes. The rate of browning was found higher in mature sago palm than the immature sago palm as indicated by the reduction of phenolic compound concentrations and development of colour during the reaction. As for the enzyme activity, the LPPO markedly decreased with maturation. This was found to be due to natural solubilization of the LPPO during maturation, thus triggered higher oxidation rate of the naturally occurring substrates present in the mature sago logs. Extreme conditions of pH and temperature, which occurred during the actual processing of sago starch significantly, effected the starch quality as well.

Conclusions

Brown discoloration of sago logs is the result of the action of two browning enzymes; LPPO and LPOD. Both enzymes, designated as LPPO1 and LPPOII and LPOD1 and LPODII were isolated and purified to homogeneity. The enzymes strongly oxidized the phenolic compounds, which are of widespread occurrence in the sago logs, in the following order: 4-methylcatechol, epicatechin, catechol and chlorogenic acid. A 20-mM concentration of *p*-coumaric acid irreversibly inhibited both the LPPO and POD.

Benefits from the study

The results obtained from this project have enabled effective preventive measures be developed for preventing the enzymatic browning reaction occurred in sago logs and sago starch during processing and storage. This method allows controlling the enzymatic browning without any effect on the starch quality

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