Ferulic acid production from wheat bran using *Staphylococcus aureus*

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Abstract: Work has been carried out to study the isolation of ferulic acid from wheat bran using *Staphylococcus aureus*. The ferulic acid was identified and quantified by HPLC. It was confirmed that about 275 mg of ferulic acid was obtained from 1kg of wheat bran after 6th days of incubation period. [New York Science Journal. 2010;3(4):79-81]. (ISSN: 1554-0200]

Key words: *Staphylococcus,* ferulic acid, wheat bran, vanillin.

1. Introduction

The yearly accumulation of agro-industrial waste-materials generated by the milling, brewing and various food based industries, has led to the consideration of extracting high value residues to offset the cost of treating and disposing of the residues. Most of these by-products are currently used as animal feed, but attention towards obtaining of high-value compounds from these wastes is being emphasized by various academic as well as industrial researchers. Biological degradation, for both economic and ecological reasons, has become an increasingly popular alternative for the treatment of agricultural, industrial, organic as well as toxic wastes. A high proportion of this waste material is carbohydrate and phenolic in nature. Specific sugar residues can be released from the cell-wall by the action of carbohydrases. On the other hand phenolics moieties are linked to polysaccharides by ester-bond forming structural cross-linking bridges between various polymers. Some phenolic compounds, such as ferulic and p-coumaric acids, are also present in cell wall Ferulic acid is the most abundant hydroxycinnamic acid in the plant kingdom and occurs mainly in the cell wall of cereal plants, which is covalently linked to lignin with ether bonds (MacAdam & Grabba 2002). Ferulic acid is a very important component for the structure and the biology of cell-wall as it can cross link polysaccharide chains through dimerisation reaction (Ishii, 1997). It is the precursor of coniferyl alcohol, a monolignol which contributes to lignification in the conifers and angiosperms. It is mainly conjugated with mono- and oligosaccharides, polyamines, lipids and polysaccharides and seldom occurs in a free state in plants. Ferulic acid has been reported to have many physiological functions including antioxidant (Graf, 1992), antimicrobial, anti-inflammatory, anti-thrombosis, anti-cancer activities and antibiotic properties (Beschia et al. 1982).

2. Materials and Methods

Microorganism

Staphylococcus aureus was isolated from soil and screened for its ability to grow in ferulic acid containing medium in the Department of Botany, Ravenshaw University, Cuttack. Pure cultures of these strains were maintained on a mixed medium containing both beef extract and peptone and cultures were incubated at 37 ^{0}C. In order to obtain high-density cultures, the bacterium was grown in a broth medium containing both beef extract and peptone (pH 7.2) for 5 days.

Agro-industrial residues (wheat bran)

Wheat bran samples were obtained from common soft wheat (*T. aestivum*) cultivars grown in local area for flour production. These materials are normally used as animal feed. The brans were obtained from nearby market in Ravenshaw University. The brans contained 0.16 % of starch (dry matter). Samples were stored at - 4 °C.

Destarching of wheat bran.

Wheat bran obtained from market, contained 0.16% of starch (dry matter). Before using as substrates, wheat bran was destarched. The procedure of destarching was described as follows. Certain amount of wheat bran was washed with tap water many times. The bran was recovered by filtration through a glass filter and the water treatment was repeated along with 1.5 % Tween 80. The processed wheat bran was soaked into filter paper and dried overnight. Again it was washed with tap water along with 1.5% Tween 80. Then it was dried in oven and autoclaved for 20 minutes at 121 °C to inactivate any

endogenous enzymes and proteinaceous inhibitors and stored at -20 °C.

Release of ferulic acid

The microorganism was grown in minimal medium (Muheim & Lerch 1999) containing wheat bran as sole carbon source. The initial pH of the minimal medium was adjusted to 7.0 before autoclaving for 15 min at 121 °C. The cultures were incubated at 35 °C and analyses were carried out in duplicates on day basis analyses upto 10 days of incubation.

Analysis of ferulic acid

Culture supernatants were prepared by filtration process. These were acidified (pH 1-2) and extracted with equal volume of ethyl acetate. The ethyl acetate was evaporated using the rotary vacuum evaporator and residue was dissolved in 50% methanol and applied to thin layer chromatography.

Separation and identification of ferulic acid

Separation and identification of ferulic acid was carried out by HPLC method. Separation of ferulic acid was performed by using HPLC with a linear isocratic solvent system. The identification of ferulic acid was confirmed by comparing retention times of external standards.

3. Results

Staphylococcus aureus was inoculated in minimal medium containing de-starched wheat bran and incubated for 10 days at 35° C and results obtained are shown in Table 1 and Fig 1.

Table 1. Release of ferulic acid from wheat bran by *Staphylococcus aureus*

Period of incubation (Days)	Amount of ferulic acid (mg/kg of bran)
1	48
2	72
3	95
4	145
5	192
6	244
7	275
8	112
9	42
10	18

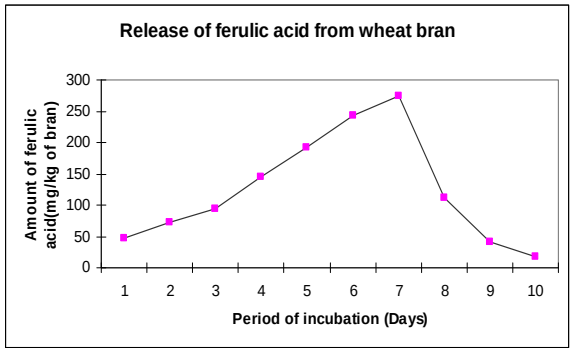

Fig. 1. Release of ferulic acid from wheat bran by *Staphylococcus aureus*

4. Discussion

Wheat bran is a major milling by – product and represents an abundant but under –exploited renewable resource. Starch depleted bran from wheat (*Triticum aestivum)* is rich in arabinoxylans (AX). Enzymatic upgrading of bran is an attractive alternative to environmentally damaging chemical methods currently used for lignocellulose saccharification. Over the last decade concentration on the isolation and purification of number of microbial esterase which can cleave ferulic acid from sugar residues in agro-industrial waste was emphasized. Wheat bran obtained from market, contained 0.16% of total sugar (dry matter). Before using as substrates, wheat bran was destarched for easy release of ferulic acid during esterase activity. In this case study, it is seen that maximum amount of ferulic acid was found to be 275mg per kg of wheat bran only after 6 days of incubation.

5. References

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