Feruloyl esterases: a tool for biotransformation of hemicellulose to bioactive compounds

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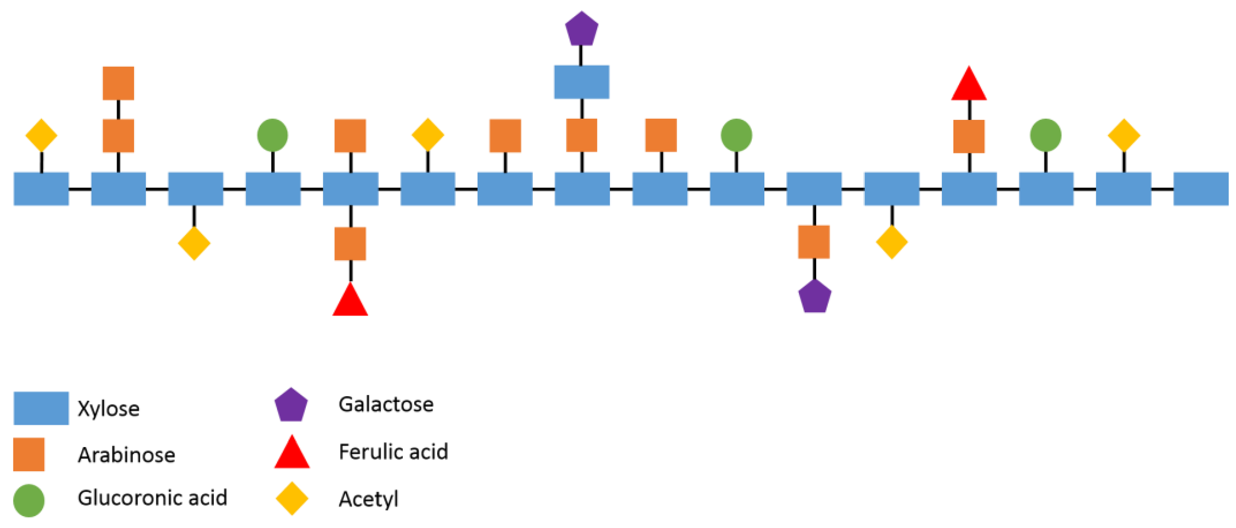
FAEs can be used to hydrolyze ferulic acid (FA) from lignocellulosic biomass, therefore helping in its depolymerization, but also in modifying FA and other hydroxycinnamic acids to produce bioactive compounds through transesterification and esterification reactions.

Lignocellulose and ferulic acid

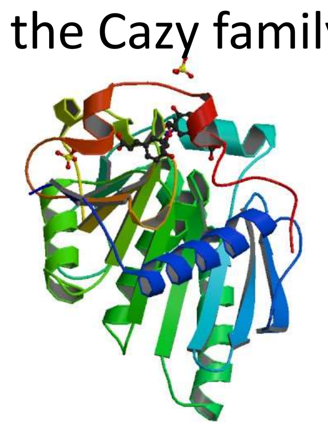
Ferulic acid is found mostly as a substituent of hemicellulose polymers. It has also been shown that diferulic acid cross linking can happen between lignin and hemicellulose [1].



Schematic representation of Xylan, one of the most abundant hemicellulose polymer.

Feruloyl esterases (FAEs)

They are a class of enzymes active on ester bounds and release FA and other hydroxycinnamic acids. They are therefore part of the hemicellulose group and classified under the Cazy family CE1.

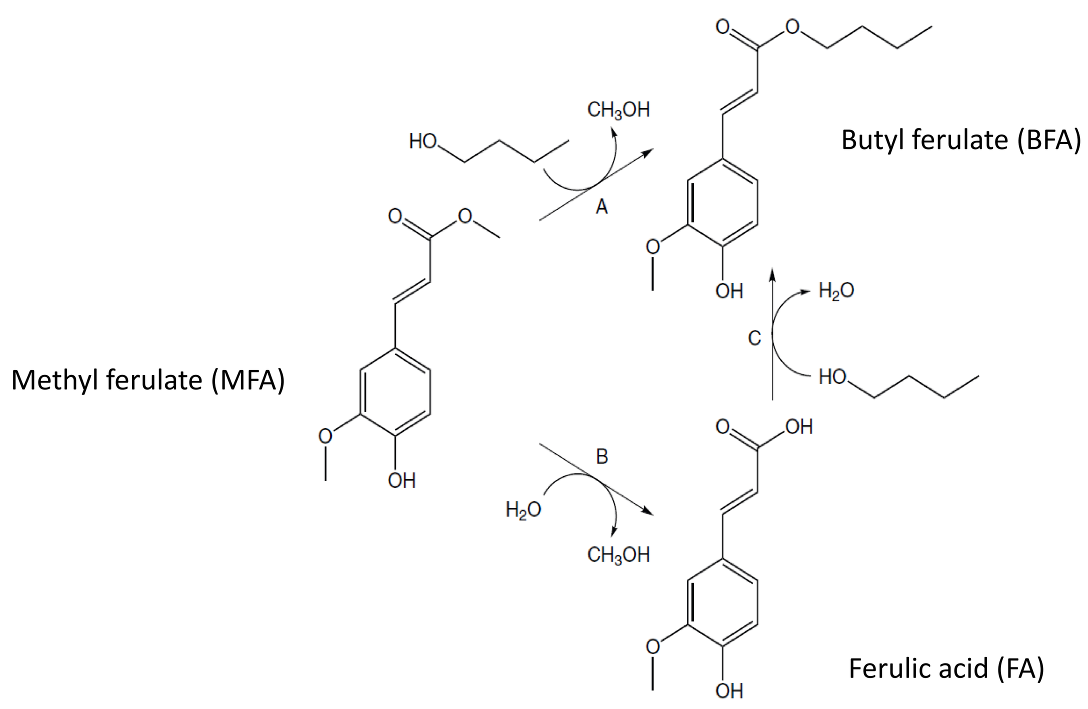


Aspergillus niger FaeA

Apart from their natural hydrolytic reaction, they can also catalyze esterification and transesterification reactions.

✓ Two new FAEs expressed

Some reactions catalyzed by FAEs

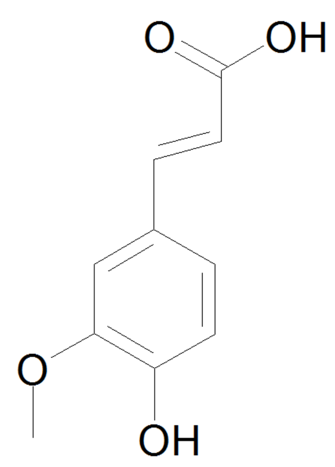


FAEs can catalyze different reactions: (A) Transesterification of MFA with 1-butanol generating BFA and methanol. (B) Hydrolysis of MFA generating ferulic acid and methanol (natural reaction at high water contents). (C) Esterification of FA with 1-butanol generating BFA and water [4].

Properties of hydroxicinnamic acids

The most known and studied of the hydroxycinnamic acids (HCA) is ferulic acid.

- Antioxidant
- Antidiabetic
- Anticancer
- Anti hypertensive
- Approved as food additive (USA, Korea, Japan) [2]



Ferulic acid (FA)

Other HCAs have been found to also have antioxidant properties [3].

FA is hydrophilic. Making it more lipophilic can be done by esterification or transesterification with FAEs.

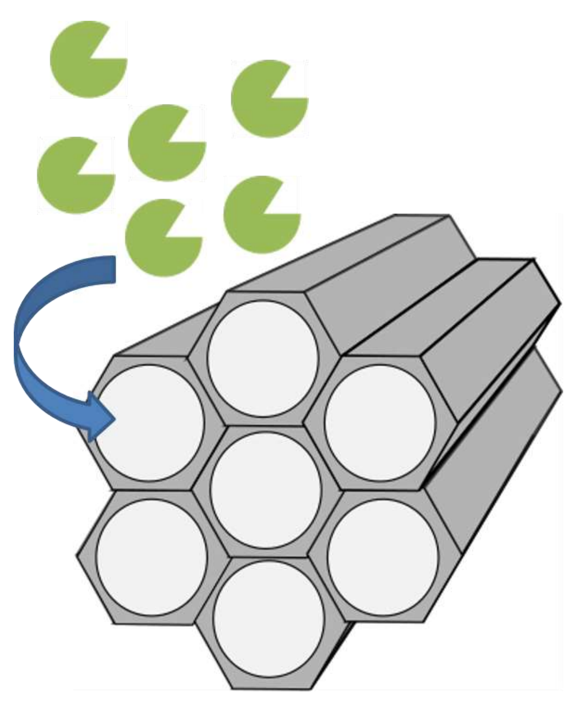
Immobilization and mesoporous silica materials

Why immobilizing?

- Reusability
- Stability
- Selectivity
- Downstream purification

Why MPS?

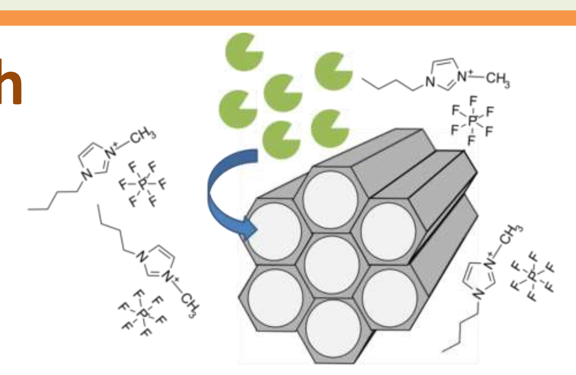
- Adsorption
- Reduces leakage
- Tunable
- Stable

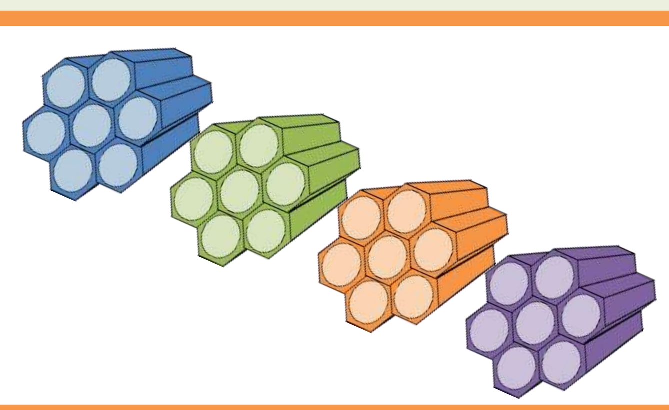


- ✓ Selectivity ratio between transesterification and hydrolysis changes upon immobilization [4]
- ✓ Immobilization in SBA-15 yields good operational stability and reusability [4]
- ✓ Immobilization yields and the following enzymatic reactions are pH dependent [5]
- ✓ Methodology to measure pH inside the pores [6]
- ✓ Methodology to follow immobilization in real time [7]
- ✓ Km unchanged upon immobilization [8]

Future research

- ✓ Effects of ionic liquids on transesterification with immobilized FAEs
- ✓ Effects of glycosylation on immobilization
- ✓ Effects of support surface modifications





[1] De Vries, R.P., and Visser, J. (2001). Aspergillus Enzymes Involved in Degradation of Plant Cell Wall Polysaccharides. Microbiol. Mol. Biol. Rev. *65*, 497–522.
[2] Teixeira, J., Gaspar, A., Garrido, E.M., Garrido, J., and Borges, F. (2013). Hydroxycinnamic Acid Antioxidants: An Electrochemical Overview. BioMed Res. Int. *2013*, e251754.
[3] Shahidi, F., and Chandrasekara, A. (2009). Hydroxycinnamates and their in vitro and in vivo antioxidant activities. Phytochem. Rev. *9*, 147–170.
[4] Thörn, C., Gustafsson, H., and Olsson, L. (2011). Immobilization of feruloyl esterases in mesoporous materials leads to improved transesterification yield. J. Mol. Catal. B Enzym. *72*, 57–64.
[5] Thörn, C., Udatha, D.B.R.K.G., Zhou, H., Christakopoulos, P., Topakas, E., and Olsson, L. (2013). Understanding the pH-dependent immobilization efficacy of feruloyl esterase-C on mesoporous silica and its structure–activity changes. J. Mol. Catal. B Enzym. *93*, 65-72.
[6] Thörn, C., Carlsson, N., Gustafsson, H., Holmberg, K., Åkerman, B., and Olsson, L. (2013). A method to measure pH inside mesoporous particles using protein-bound SNARF1 fluorescent probe. Microporous Mesoporous Mater. *165*, 240–246.
[7] Thörn, C., Gustafsson, H., and Olsson, L. (2013). QCM-D as a method for monitoring enzyme immobilization in mesoporous silica particles. Microporous Mesoporous Mater. *176*, 71–77.
[8] Bonzom,C., Schild, L., Gustafsson, H., and Olsson, L. *Manuscript in preparation*. Immobilization of a feruloyl esterase in mesoporous silica particles and its biochemical characterization in hydrolysis and transesterification reactions.