

# Can ultrasonic waves improve enzymolysis for biomedical applications?

## Abstract

In recent years, ultrasound (US) has been successfully used in a large number of crucial bioprocesses such as upregulation of enzymatic hydrolysis (biocatalysis). Ultrasound treatment or pretreatment are supposed to activate/accelerate enzyme catalysis enhancing product formation. Such methods might offer high efficiency of enzymatic bioconversion and production of new biologically active peptides. We present here a short overview of the possible effects of ultrasound exposure to improve enzymatic processes and to minimize operational costs in biotechnological applications aiming to biomedical field.

**Keywords:** biocatalysis, accelerate, upregulation, bioconversion, enzymatic,  $\alpha$ -amylase, immobilized, diffusion

Volume 4 Issue 5 - 2017

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**Received:** April 22, 2017 | **Published:** May 01, 2017

## Introduction

Acoustic cavitation has long been reported as the main responsible for changes in the physical properties of a liquid medium exposed to ultrasonic waves, giving rise to the formation of collapsing bubbles which lead to small pressure and temperature alterations. Since enzyme-substrate interactions are naturally favored by the high molecular diffusion rates in liquids, consequent changes in free energy of the system would directly alter the catalytic potential of the reactions.<sup>1</sup>

On the other hand, the diffusion of substrates and direct mechanical forces are also reported for alterations in the enzyme conformation, which leads to enzyme activation.<sup>2</sup>

The discussion of the use of US exposure as tool for improving biocatalysis is providential to the application in biotechnological processes. This study aims therefore to provide an insight of the possibilities that the US effects on enzymolysis offers to further researches and biotechnological processes.<sup>3</sup>

## Effect of ultrasound on the activity and conformation of enzymes

Factors such molecular alterations and mechanical forces from the direct action of ultrasonic waves would also be responsible for improvements on the enzymolysis. Researches have verified that the ultrasound exposure affected contrarily on the secondary and tertiary structures of pepsin and  $\alpha$ -amylase, with enhanced and inhibited activity respectively.<sup>4</sup> The author discussed that direct action of the ultrasonic waves on the enzyme structure lead to conformations changes that could activate or inactivate the enzymes.<sup>5</sup> That brings the idea that the enzyme structure is crucial to determine whether an enhancement will be provided.

On the other hand, the activity of immobilized enzymes have been described as affected by US exposure for changes in the concentration gradient and diffusion constants of the substrate and such activities presented increases in the reaction rate of 200%.<sup>6</sup>

## Enhanced production of bioactive products by ultrasound stimulation

Bashari et al.<sup>7</sup> discuss that ultrasonic irradiation was used to improve the enzyme activity both by isolated action or combined with other agents: they verified that the kinetic activity of dextranase and the hydrolysis rate were improved and that the combined action of high frequency ultrasounds with high hydrostatic pressure enhances the enzymatic hydrolysis of dextran catalyzed by dextranase. Challenges associated with the introduction of new peptide products, for example, include proteolytic degradation methods like US, fast clearance in the body, low solubility in water, immunogenicity and regulatory hurdles.<sup>8</sup>

## Applications of the US as tool for biocatalysis

Activities of fibrinolytic enzymes,  $\alpha$ -amylase, pepsin, dextranase and other enzymes are reported as enhanced under US exposure.<sup>9,10</sup> Thus the applications of the US devices in clotting processes, food industry are interesting for biotechnological applications. Besides, have studied the US effect upon the process of proteolytic autoactivation of the serine proteases precursors—chymotrypsinogen and trypsinogen. Ultrasound has also proposed as an economically feasible pretreatment alternative.<sup>11,12</sup> The effects of ultrasound on sludge includes particle size reduction, organic matter solubilization, enzyme release and stimulation of biological activity.<sup>13</sup>

## Conclusion

Ultrasound exposure is able to enhance enzyme reactions, although particular molecular conformations and conditions of exposure are required. Methods for improvement of the enzyme activity are highly suggested for further researches, included possible screening for enzymes activated by ultrasound exposure.

## Acknowledgements

The authors are thankful for the financial support from FACEPE (Foundation for Science and Technology of the State of Pernambuco)

and CNPq (National Council for Scientific and Technological Development).

## Funding

None.

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