Keywords: Dagaa; Optimization; hydrolysis; Alcalase

Introduction

From previous proximate composition analysis studies of Dagaa, we have established that it is a nutritionally dense sh [1]. In addition, it has the second highest landings (of up to 63%) in Lake Victoria. However, poor storage and handling conditions have led to postharvest losses of 30% which increase to 50% during the rainy season. Alternative methods of using this under-utilized Dagaa sh species are therefore needed to increase its potential utilization and market value [2].

Use of enzymes to recover sh protein hydrolysates (FPH) is a technology that is gaining popularity due to the bioactive properties associated with these compounds. Enzymatic hydrolysis of native sh proteins has been shown to improve their functional properties, including solubility, emulsifying capacity and foaming characteristics hence o ering interesting opportunities for food applications and pharmaceuticals [2].

In any given enzymatic process, the nal product yield depends on several factors. ese include; the type of enzyme and substrate, hydrolysis conditions; pH, temperature, time and enzyme/substrate ratio, solvent ratio and stirring speeds [3-6].

In addition to e ect on yield, enzyme type also a ects the bioactivity of the obtained hydrolysate. erefore, appropriate selection of suitable enzyme and substrate as well as hydrolysis conditions such as; enzyme to substrate ratio, hydrolysis time, pH and temperature are crucial in obtaining protein hydrolysates with desirable functional and biological properties [5]. Moreover, from an economical point of view, the amount of enzyme used should be optimized to prevent enzyme waste and manage its costs [6].

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properties of the Alcalase hydrolysate were more potent than that of hydrolysates produced by the endogenous Dagaa enzymes [2].

Stirring

For this study, optimised stirring for Dagaa (*Rastrineobola argentea*) was xed at minimum stirring speed for an overhead stirrer (Stuart, UK). is was the point at which the substrate was seen to move as one (without a broken meniscus) re ecting optimum mixing. On the other hand, higher speeds (500-2000 rpm) caused spattering indicative of poor mixing.

Optimization for solvent ratio

Solvent plays an important role in enzyme processes. is is important for the type of solvent as well as the amount of solvent. is is because the solvent components have an e ect on hydrolysate components and the stability of the enzyme used. In this study, there was a negative correlation between solvent ratios and percent nitrogen recovery. Hence an increase in percent solvent ratio ((v/w)) led to decrease in percent nitrogen recovery (Figure 1). e maximum percent nitrogen recovery was obtained at solvent ratio 0.5% (v/w). is was considered to be the optimum solvent ratio.

is could be explained by the dilution. High solvent ratio dilutes the product, whereas low solvent ratios result in concentration of the substrate thus low activity demonstrated by minimised low percent nitrogen recovery. A decrease or increase in solvent ratio above the optimum reduces the yield (%NR). Similar results have been obtained for lipase where increasing solvent ratio resulted in decreased product yields [27].

Optimization for pH

Previous studies on other substrates have shown Alcalase to have activity within the alkaline pH range. e pH pro le for the hydrolysis of

Optimization for enzyme/substrate ratio

Apart from pH and temperature, enzyme substrate ratio is one of the factor, which shows a marked in uence on peptide bond cleavage of the protein substrate [32]. e pro le of % NR during the 2 h hydrolysis of Dagaa is shown in Figures 4. Maximum %NR was obtained at ES ratio of 2% (v/w).

Approximately 20% of the total nitrogen remained insoluble at the end of a hydrolysis process even if more enzymes were added during the stationary phase of hydrolysis [35]. is insoluble residue contains peptides enriched with hydrophobic amino acids that are highly resistant to further degradation by the enzyme [33]. Furthermore, the increase in peptide concentration in the hydrolysis mixture and

production of protein hydrolysate with antioxidative function, it was a requirement to stop the proteolysis at least at the dipeptide stage.

Consequently, in this study, optimized hydrolysis conditions were xed at minimum stirring speed (overhead Stuart stirrer, UK), 56°C, pH 7, ES ratio of 2% (v/w) and solvent ratio of 0.5 (v/w) for 6 hours. Following continuous hydrolysis at these optimum conditions, an 83% degree of hydrolysis was achieved with a 71% protein yield.

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