I. INTRODUCTION

1.1 Background

Enzymes are biological compounds that serve as catalysts, speeding up biochemical and biological reactions (Panneerselvam, 2015). Without the use of a catalyst, an enzyme can accelerate a reaction 108 to 1011 times faster (Poedjiadi, 2006). Protease enzymes are one class of enzymes that is crucial to the expansion of the industry. Around 75% of enzyme applications sold globally for industrial use are hydrolytic enzymes, and roughly 60% of those are proteolytic enzymes (Ningthoujam& Kshetri, 2010; Rai *et al*., 2010).

Proteases are enzymes that catalyze the breaking of peptide bonds within proteins and polypeptides, producing oligopeptides or free amino acids as a result. Proteinases, which catalyze the degradation of protein molecules into more manageable fragments, and peptidases, which hydrolyze polypeptide fragments into amino acids, are two types of protease enzymes. Every live cell produces proteases for intracellular use or to be released into the environment for nutritional and defensive purposes. Aside from being physically necessary, they have also found various crucial functions to play in the health, food, textile, and medicine industries over the years (Correa *et al*., 2014).

Proteases, also known as proteolytic enzymes, are commercially important because they are used in bioremediation and waste treatment, detergents, cosmetics, and leather manufacture, silk degumming, animal cell culture, contact lens cleaning, therapy and diagnosis, and the pharmaceutical, photographic, and food industries (Rao *et al.*, 1998). Accordingly, many proteases are being studied by the

pharmaceutical industry as possible therapeutic targets or as diagnostic and prognostic agent biomarkers (Turk, 2006). Proteases also serve important functions in plants, contributing to the processing, maturation, or destruction of certain protein sets in response to developmental cues or changes in environmental conditions. Furthermore, proteases are thought to be insecticides because they are required for the complete digestion of complex insect cuticles (Anwar & Saleemuddin, 1998; Gupta *et al.*, 2002; Harrison & Bonning, 2010; Hasan *et al.*, 2013; Kumar & Takagi, 1999; Murthy & Naidu, 2010; Nielsen & Oxenboll, 1998).

Protease activity, like that of most enzymes, is controlled by a variety of variables. Temperature, pH, and enzyme activators are among the most influential parameters (Leboffe, 2012). The majority of processes industry requires enzymes that are able to withstand extreme conditions. So it is very important for enzymes used to have optimal conditions in the range a wide range of temperatures and pH (Bizuye, 2014). The appropriate incubation period will have resulted in maximum protease production with the resulting high enzyme activity (Yuniati, 2015).

Plants, animals, and microbes all have the ability to create proteases. Microorganisms are a source of very beneficial enzymes because these microorganisms have faster growth than animals and plants (Yuliana & Nuniek, 2014). Due to their lower production costs and greater stability, microbial enzymes are among the most significant of them. Since microbes can manufacture such a wide range of enzymes, their ability to produce proteases has increased recently. According to Tavano (2013), they produce nearly two-thirds of the proteases produced worldwide. Since fungi produce a wider variety of enzymes than bacteria

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do, and mycelia are easier to separate from the culture medium, fungi have increased their production of proteases in recent years (Veloorvalappil et al., 2013; Haddar et al., 2010). Plants can act as a reservoir for a variety of microorganisms known as endophytes (Bacon *et al.*, 2000).

Endophytes are microorganisms that live inside their hosts' tissues and carry out ecological functions without endangering the host. They are the source of numerous novel biomolecules, such as enzymes, and have been found all over nature (Pinheiro *et al.*, 2012). Endophytic fungi are intracellular live organisms in healthy plant tissues, which induce the host to produce secondary metabolites. This induction can be caused by genetic recombination or coevolution (Sugijanto *et al.*, 2004; Sia *et al*., 2013). Huang in White *et al* (2014) stated that there is a correlation between the presence of endophytic fungi and the host plant's ability to produce secondary metabolites. The ability of endophytic fungi to synthesize secondary metabolites is an opportunity for large-scale production in a short time without causing ecological damage. The fungi found in mangrove endophytes have a significant potential to create secondary metabolites. The leaves of the mangrove plants serve as a habitat for many kinds of microfungi. Endophytic fungi play a crucial part in the process of plant nutrient uptake (Chanway, 1996).

Biotechnology Laboratory Andalas University has collections of endophytic fungal isolates from *sonneratia alba* namely EUA-120, EUA-121, EUA-122, EUA-123, EUA-124, EUA- 125, EUA-126, and EUA-127, and two isolates positive protease-producing with the codes EUA-124 and EUA-126. Based on the enormous ability of endophytic fungi and the biodiversity that exists in Indonesia, the prospect

of research on endophytic fungi from plants in Indonesia is very large. Mangrove areas are the main ecosystems that support important life in coastal and marine areas. Many benefits can be taken from mangrove plants (Saprudin & Halidah, 2012). Considering its potential as a mangrove plant that lives in the tropics is very large, it is important to conduct research on endophytic fungi of mangrove plants as a producer of protease enzymes. This study aims to analyze and explore the optimum condition of protease production by endophytic fungi of mangrove plants.

1.2 Problem Formulation

Based on the description above, the formulation of problems in this research were:

1. what is the optimum temperature and pH for two endophytic fungal isolates of mangrove plants *Sonneratia alba* from Mandeh area, Pesisir Selatan District to produce protease?

2. How is the enzyme production after optimization?

1.3 Research Objective

The objectives of the research were:

1. To determine the optimum temperature and pH of endophytic fungal isolates of mangrove plants *Sonneratia alba* from Mandeh area, Pesisir Selatan District in producing protease.

2. To determine the enzyme production after optimization.

1.4 Research Benefit

The research benefit provide scientific information about the optimal temperature and pH for the production of protease enzymes and enzyme production after

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optimization so that it can be used to increase the production of protease enzyme from endophytic fungi that can be applied to various industries.

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