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Design of a gold nanoprobe for the detection of *Pseudomonas aeruginosa* elastase gene (lasB)†

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Nosocomial infections are one of the major health problems that increase mortality. *Pseudomonas aeruginosa* is one of the important causes of nosocomial infections. This bacterium has a gene called the lasB gene, the product of which is a zinc-dependent metalloprotease. This gene plays a central role in the pathogenesis of *Pseudomonas* spp. This pathogen is highly toxic and destroys tissues and also has a moderating effect on the immune system; on the other hand, it initiates the intracellular pathway of biofilm growth. Although there are methods such as molecular methods for identifying the lasB gene, due to the high cost and the need for specialized personnel, it is necessary to replace them with an appropriate method. In this study, a gold nanoparticle-based DNA diagnostic sensor sensitive to the aggregation states of gold nanoparticles was used to identify amplified and non-amplified lasB genes. The results of the experiment were evaluated both visually and spectrally. The minimum detection value of this method was 10 ng of the amplified lasB gene and 50 ng of the non-amplified lasB gene. This method is very fast, simple, easy and low cost.

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1 Introduction

Nosocomial infections have been one of the major health problems for a long time, and they increase the mortality rate in hospitalized patients. These infections have increased health care costs.[1] About 5% to 10% of hospitalized patients are infected with a nosocomial infection.[2] According to WHO studies, the prevalence of nosocomial infections in developing countries is between 5.7% and 19/1%, while it is 7/6% in developed countries.[3] According to the studies in Iran, the most common bacteria that are isolated from patients with nosocomial infections include *Pseudomonas aeruginosa* (24/3%), *Klebsiella pneumoniae* (18/6%) and enterobacteria (14/3%).[1] The genus *Pseudomonas* belongs to the family of Pseudomonadaceae and to the class of Pseudomonadales. *Pseudomonas aeruginosa* is a ubiquitous Gram-negative opportunistic bacterium,[4] which is the most important cause of nosocomial infections in patients with weakened immune systems; *Pseudomonas* infections have led to a mortality rate of about 50% in these patients.[5–7] *Pseudomonas aeruginosa* is one of the most common causes of lower respiratory tract infection. It has a gene called lasB that produces a zinc-dependent metalloprotease called *Pseudomonas* elastase or pseudolysin that plays a critical role in the pathogenesis of *Pseudomonas aeruginosa*. This pathogen is highly toxic and damages tissues and also has a moderating effect on the immune system; on the other hand, it initiates the intracellular pathway of biofilm growth. These are the pathogenic mechanisms of lasB that cause a chronic infection to develop.[8] To identify *Pseudomonas aeruginosa* bacteria, gram staining and culture in different environments can be used, which are time consuming and do not have high sensitivity and specificity.[9,10] lasB can be identified using other methods, such as PCR and real-time PCR. These two methods, although highly specific and sensitive, have limitations due to the requirement of dedicated reagents, expensive equipment and specialized personnel.[11,12]

Recently, researchers have shown great interest in using gold nanoparticles to identify pathogens.[13–20] This method of detection is based on the property of surface plasmon resonance (SPR) of gold nanoparticles. The change in the dispersion state of the nanoparticles results in a color change, which results in detection.[21,22] Many studies have used gold nanoparticles containing thiol-modified oligonucleotides (Au nanoprobes) for diagnosis.[23] These nanoparticles have unique optical properties. These features enable us to visualize their color change and thus, no advanced tools are needed in this method. These nanoprobes can be used in two ways to identify nucleic acid sequences using a cross-linking method (CL) and a non-cross-linking method (NCL).[21,23–25] In the NCL colorimetric assay, attaching the complementary portion of the target molecule to the nanoprobe makes the nanoprobes resistant to salt-induced

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