Introduction: Gold nanoparticle (Au) biosensors, tiny particles that can appear red or blue, play a crucial role in rapid diagnostic tests for a variety of diseases. This study explores how the color and size of these particles affect the effectiveness of the tests. The use of red-colored AuNPs (AuNP-R) is common in diagnostic kits due to their complex production process and robust structure. Our research focuses on comparing these with blue-colored AuNPs (AuNP-B), which are characterized by a different particle size and a simplified production process.

Objectives: Our goal is to evaluate and compare the effectiveness of blue and red gold nanoparticles when conjugated with Protein A in rapid diagnostic tests.

Methodology: We synthesized the AuNPs in our lab using a modified Turkevich method, followed by characterization through ultraviolet-visible spectroscopy, dynamic light scattering, laser doppler electrophoresis, and transmission electron microscopy. The blue and red AuNP bioconjugates were prepared in a stabilizing buffer (pH 8.0), ensuring both had the same optical density (OD=50). We then assessed their effectiveness in detecting Canine Visceral Leishmaniasis via a lateral-flow rapid diagnostic test, using 90 dog serum samples (30 positive, 60 negative) to compare their performance. Kappa’s statistical coefficient was calculated with GraphPad.

Results: Among 30 positive samples, AuNP-R presented one false-negative sample, while AuNP-B obtained four false-negatives, resulting in sensitivities of 96% and 86%, respectively. In negative samples, two false positives were obtained utilizing both bioconjugates amongst (specificity = 97%). The intensity of the test line was also compared between bioconjugates and a decreased signal in blue compared to red bioconjugate was observed. The control line intensity was similar for both bioconjugates. Kappa’s coefficient (0.924) indicated almost perfect agreement between results in both colored tests.

Conclusion: Blue gold nanoparticles (AuNP-B) have shown promise for use in rapid diagnostic tests, although adjustments are needed to improve signal intensity. Future research should focus on optimizing these nanoparticles for various diseases and evaluating their long-term stability.

Keywords: Gold Nanoparticles; Protein A; Rapid Diagnostic Tests