INTISARI

Penelitian ini bertujuan mengoptimalkan ekstraksi double-stranded RNA (dsRNA) dari tanaman terinfeksi virus menggunakan konsentrasi etanol 14%, 16%, dan 18% dalam *buffer* STE 1× pada jaringan tanaman tembakau yang terinfeksi *Tobamovirus*, tomat terinfeksi *Cucumber mosaic virus* (CMV), dan bawang merah terinfeksi *Potyvirus*. Tujuan spesifik mencakup identifikasi pengaruh etanol terhadap kemurnian dan hasil dsRNA, penentuan konsentrasi optimal berdasarkan jenis virus dan tanaman, serta evaluasi efektivitas deteksi RT-PCR. Metode penelitian meliputi pengumpulan sampel dari Magelang, Bantul (Yogyakarta), homogenisasi jaringan, adsorpsi dsRNA pada selulosa CF-11, presipitasi etanol, kuantifikasi NanoDrop (A260/A280, A260/A230), RT-PCR dengan primer spesifik (TobamodF/R, CMV-RNA3F/R, MJ1/MJ2), elektroforesis, sekuensing, dan analisis BLAST/MEGA12. Semua ekstraksi RNA berhasil diamplifikasi dengan RT-PCR (880 bp untuk *Tobamovirus*, 540 bp untuk CMV, dan 327 bp untuk *Potyvirus*). Hasil kuantifikasi menunjukkan konsentrasi etanol optimal bervariasi, Konsentrasi etanol optimal untuk ekstraksi dsRNA bervariasi, yaitu 16% pada tembakau (4,48 ng/µL), 14% pada tomat (34,55 ng/µL), dan 18% pada bawang merah (9,27 ng/µL). Kemurnian keseluruhan baik, dibandingkan kit komersial.

Kata kunci: ekstraksi dsRNA, konsentrasi etanol, RT-PCR, selulosa, virus tanaman.

ABSTRACT

This study aimed to optimize the extraction of *double-stranded* RNA (dsRNA) from virus-infected plants using ethanol concentrations of 14%, 16%, and 18% in 1× STE buffer, applied to tobacco infected with *Tobamovirus*, tomato infected with *Cucumber mosaic virus* (CMV), and shallot infected with *Potyvirus*. Specific objectives included identifying the effect of ethanol concentration on dsRNA purity and yield, determining the optimal concentration based on virus and plant type, and evaluating the effectiveness of RT-PCR detection. Research methods involved sample collection from Magelang and Bantul (Yogyakarta), tissue homogenization, dsRNA adsorption onto CF-11 cellulose, ethanol precipitation, NanoDrop quantification (A260/A280, A260/A230), RT-PCR using specific primers (TobamodF/R, CMV-RNA3F/R, MJ1/MJ2), electrophoresis, sequencing, and BLAST/MEGA12 analysis. All RNA extractions were successfully amplified by RT-PCR, yielding bands of 880 bp for *Tobamovirus*, 540 bp for CMV, and 327 bp for *Potyvirus*. Quantification results indicated that the optimal ethanol concentration varied by plant matrix: 16% for tobacco (4.48 ng/µL), 14% for tomato (34.55 ng/µL), and 18% for shallot (9.27 ng/µL). Overall purity was satisfactory, compared to commercial kits.

Keywords: dsRNA extraction, ethanol concentration, RT-PCR, cellulose, plant virus.