Total RNA extraction from the Aromatic Phalaenopsis bellina, Endemic in Sabah, Borneo

ABSTRACT

Phalaenopsis bellina is an attractive orchid due to its unique appearance and distinctive floral fragrance. Many past studies on this plant had focused on the plant at the molecular level; however, it requires sufficient quantities of high-quality P. bellina RNA. RNA is more delicate to manipulate than DNA due to its structural instability and its vulnerability to various secondary metabolites, such as polyphenols and polysaccharides. Therefore, in this study, four RNA isolation methods which are a modified phenol-chloroform method and three commercial kits (Vivantis, Novogene, and Analytik Jena) were used on the leaves and flowers of P. bellina for comparison. The yield and purity of total RNA were determined using spectrophotometry. The results showed that the total RNA isolated using the modified phenol-chloroform method had the highest yield (1223.75 \pm 68.51 ng/µL) and purity compared to the three commercial kits with an OD260/280 value of 2.07 and an OD260/230 value of 2.26, respectively. In particular, the isolated RNA did not show any detectable genomic DNA contamination or other impurities. The RNA isolated using the phenol-chloroform method was also evaluated by electrophoresis, reverse transcription, and PCR. The results indicated that the phenol-chloroform method appears to be superior for total RNA extraction. Thus, this developed method is proven to be suitable for the RNA extraction of plants rich in polysaccharides and polyphenols, and amenable for future molecular studies on P. bellina.