

Figure 6. Restriction analysis to confirm the construction of pITB139, pITB239-GFP and pITB260.

- A. *HindIII* digested λ DNA marker.
- **B.** Digestion of pGEMT-Easy-IRT1 with *XbaI* (lane 1) and *XbaI-SalI* (lane 2) confirmed that the *IRT1* gene was inserted into pGEMT-Easy; M: Marker HindIII λ .
- C. Digestion of pITB139 (lane 1) and pITB239 (lane 2) with *XbaI-SalI* showed a 1.0 kb band in pITB123 and a 2.7 kb band in pITB239 indicating th replacement of T7 RNAP gene in pITB239 with IRT1 gene.
- **D.** Digestion of pGEMT-Easy-GPF *HindIII* (lane 1) and *XbaI-BamHI* (lane 2) showed 1.0 kb band and 0.7 kb bands, respectively indicating the direction of cloning.
- E. Digestion of pITB239-GFP (lane 1) and pITB239 (lane 2) with *HindIII* showed a 1.0 kb band in pITB239-GFP and absent in pITB239-GFP. Digestion of pITB239 (lane 3) and pITB239-GFP (lane 4) with *BglII-SalI* showed a bigger band in pITB239-GFP (~4.5 kb) than a band in pITB239 (3.5 kb) confirming the GFP cloning.
- F. Digestion of pITB250 (lane 1) and pITB260 (lane 2) with *BglII-Smal* showed a 0.8 kb band in pITB250. Digestion of pITB250 (lane 3) and pITB260 (lane 4) with *HindIII-Smal* showed two bands in pITB250 and one band in pITB260 confirming the cloning of GFP.