



**Figure 20.** Restriction analysis to confirm the construction of pITB347 and pITb348 vectors (see figure 17. and 19 for other details).

- A. *HindIII* digested  $\lambda$  DNA marker
- B. Digestion of pA8-T7-PDS (l), pA8-T7-PDS (r) and pCAMBIA2300 (A8) with *Ncol* (lanes 1, 2 and 3) and *KpnI* (lanes 4, 5 and 6) to determine insertion and direction of "PDS promoter:T7RNAP:35S polyA" into pCAMBIA2300. Lane 1 and 4: pA8-T7-PDS(r); Lanes 2 and 5: pCAMBIA2300; Lanes 3 and 6: pA8-T7-PDS(l). Note that in lane 1 has two bands of similar size (6.1 kb and 7.6 kb).
- C. Confirmation of pA8-T7-PDS(r) (lane 1) and pITB347 (lane 2) vectors. Digestion with *BamHI-BglII* released a ~1.0 kb (35S promoter) from pA8-T7-PDS(r) and ~4.0 kb (35S promoter and T7 promoter:*uid*A:T7 terminator and T7promoter:HBsAg:T7 terminator) from pITB347.
- D. Digestion of pA8-T7-PDS(l) (lane 1) and pITB348 (lane 2) with *SalI-BamHI* that determined the cloning of "T7 promoter:*uid*A:T7 terminator and T7 promoter:HBsAg:T7 terminator" cassette into pA8-T7-PDS(l) in anticlockwise direction.