Trizol RNA extraction

1) Wash cells with 1x PBS, apply 1ml Trizol (35mm or 6-well), 3ml for 60mm, 8ml for 10cm. Pipette several times to lyse cells.
2) Incubate 5 min. at RT (room temperature)
3) Add 0.2ml chloroform to 1ml Trizol, shake vigorously for 15s, incubate 2 - 3 mins. at RT
4) Centrifuge 12K x g at 4°C, 15 mins.
5) Pipette aqueous phase into a new tube, add 0.5ml of isopropanol (per 1ml Trizol), incubate 10mins. at RT
6) Centrifuge 12K x g at 4°C, 10 mins.
7) Remove supernatant, wash 1ml 75% EtOH.
8) Vortex, centrifuge 5 mins at 4°C.
9) Air dry, resuspend in RNase-free water, dissolve 55 - 60 deg. for 15 mins.