date:

Operating Instructions

23.10.2009

Thermocycler Techne TC-3000



Hazards for Humans and Environment



Risk of $\ensuremath{\textbf{burns}}$ at $\ensuremath{\textbf{hot}}\xspace$ surfaces

Do not use next to inflamable liquids or gases. Do not put liquids directly into the unit

Rules of Behaviour

Prepare the PCR mix (on ice) into a 0.2 ml thin wall, flat cap PCR-tube Do not add the DNA-polymerase if it is not a hot-start polymerase (*i.e.* not active before initial heating) Turn on Thermocycler. Choose "Programs" and press Select ($\uparrow \downarrow$) the apropiate program (e.g. "Standard Genome") and press \downarrow to select it Use $(\uparrow \downarrow)$ to "run", "view" or "edit", ... the program. Confirm selection with \downarrow On "view" or "edit", use $(\uparrow \downarrow)$ to navigate the stages. For "edit" use \downarrow to select, type number or toggle $(\uparrow$ ↓), confirm with ↓. For the steps: digit new temp., ↓, digit min [toggle to hours with "•"], "•", digit sec, , digit ramp [0 = max.], \downarrow . Omit changes with \downarrow . Press "•" to insert or "←" to delete a step. 1) NAME 105 °C 2) Heat Lid: set lid temperature Prehead Lid: heat up lid before PCR cycles start (4 min) On pauses program for to place the PCR tube. resume " || " Pause: On 94 °C initial denaturalisation of DNA. for vector DNA less time 3) In. denat. 5m00 75 °C pauses programm for to add DNA-polymerase. resume " II " Hot start 4) Number Cycles 30 94 °C Step 1 0m30 denaturalisation of the DNA max 55 °C primer annealing: $T_m = \Sigma (\Sigma AT)^2 + (\Sigma GC)^4 [^{\circ}C]$ Step 2 0m30 max elongation: for Pfu 500 bp/min, for Tag 1,000 bp/min 72 °C Step 3 1m00 max 5) Final Extn 72 °C 5m00 final extention 10 °C Hold keeps temperature at 10 °C until program stop End programing by pressing ∇ to save and confirm changes with \downarrow (or ∇ to leave w/o changes)

For more complicate programing, refer to the manual. Use ∇ to go back to main menu.

On RUN confirm running with J. On pauses (heat lid, hot start) resume with " || "

A run can be paused and resumed by pressing " II " and can be stopped by ∇ (confirmed by \downarrow) At the end of the run (∇) confirm ending with \downarrow

Cleaning & Maintenance



After use keep the lid open to dry the heater block

Clean the heater block monthly with cutton buds soaked with 50:50 water/isopropanol