# Bench sheet · EnGeL-MOLB-PICOGREEN

**Double-stranded DNA concentration in 96-well plates with PicoGreen (FLUOstar Omega plate reader)** · version 1.1.1 (2026-11-20). Full protocol: EnGeL-MOLB-PICOGREEN. Tick each step when done; write deviations in Notes.

| | |
|---|---|
| Date | |
| Operator | |
| Plate ID / samples | |
| PicoGreen kit lot | |

**PicoGreen standards.**

| Standard | Lambda working stock (µL) | 1X TE (µL) | Working reagent (µL) | Concentration (ng/mL) |
|---|---|---|---|---|
| S1 | 50 | 0 | 50 | 1000 |
| S2 | 5 | 45 | 50 | 100 |
| S3 | 0.5 | 49.5 | 50 | 10 |
| Blank | 0 | 50 | 50 | 0 |

## Before starting

| ☐ | No. | Step |
|---|---|---|
| ☐ | • | Use filter tips at all steps; plastic tubes only. |
| ☐ | • | Keep the PicoGreen reagent and working reagent protected from light. |

## Plate and reading

| ☐ | No. | Step |
|---|---|---|
| ☐ | 1 | Pipette each standard into the black plate in duplicate (table in Reagent preparation). |
| ☐ | 2 | Samples: 2 µL sample + 48 µL 1X TE per well + 50 µL working reagent. |
| ☐ | 3 | Mix by pipetting or a short plate shake. |
| ☐ | 4 | Incubate for 2 min at room temperature, protected from light. |
| ☐ | 5 | Plate reader: *Manage Protocol* → "PicoGREEN_LC_100"; *Start measurement* → *Gain Adjustment*; then *Start measurement*. The reader protocol is a fluorescence-intensity endpoint read (Greiner 96 F-bottom plate; excitation filter 485-12, emission filter 520; top optic; 30 flashes per well; settling time 0.5 s). The gain is set at 80–90% of the highest standard (1 000 ng/mL), and the blank is subtracted in the evaluation. |
| ☐ | 6 | In the reader software, fit a linear regression of the standards (S1–S3 and blank) (*Calculation* → *Standard curve calculations* → *Linear regression fit*); then, in *Microplate View*, select *Linear Regression Fit* under *Recalculate concentration*. |

## After finishing

| ☐ | No. | Step |
|---|---|---|
| ☐ | • | Clean the bench with 70% ethanol. |
| ☐ | • | Empty the small bench bin into the right waste bin. |
| ☐ | • | Wash your hands. |
| ☐ | • | Turn off equipment and lights. |
| ☐ | • | Lock doors, fridge and freezer. |

**Notes / deviations**

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