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## Data Article

# Data on the influence of temperature on the growth of *Escherichia coli* in a minimal medium containing glucose as the sole carbon source for the joint computation of growth yields and rates at each temperature from 27 to 45°C

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## ABSTRACT

Temperature is a key factor influencing microbial growth rates and yields. In literature, the influence of temperature on growth is studied either on yields or rates but not both at the same time. Moreover, studies often report the influence of a specific set of temperatures using rich culture media containing complex ingredients (such as yeast extract) which chemical composition cannot be precisely specified. Here, we present a complete dataset for the growth of *Escherichia coli* K12 NCM3722 strain in a minimal medium containing glucose as the sole energy and carbon source for the computation of growth yields and rates

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at each temperature from 27 to 45°C. For this purpose, we monitored the growth of *E. coli* by automated optical density (OD) measurements in a thermostated microplate reader. At each temperature full OD curves were reported for 28 to 40 microbial cultures growing in parallel wells. Additionally, a correlation was established between OD values and the dry mass of *E. coli* cultures. For that, 21 dilutions were prepared from triplicate cultures and optical density was measured in parallel with the microplate reader (OD<sub>microplate</sub>) and a UV-Vis spectrophotometer (OD<sub>UV-vis</sub>) and correlated to duplicate dry biomass measurements. The correlation was used to compute growth yields in terms of dry biomass.

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Specifications Table

Subject	Microbiology: General
Specific subject area	Data describes OD and dry mass measurement of <i>E. coli</i> culture growth in minimal media for different temperatures. The data was used to calculate growth rates and growth yields.
Type of data	Table, Figure, Raw data, Filtered data
How the data were acquired	Overnight cultures of <i>E. coli</i> grown in modified MOPS media were used for analyses. A thermostated microplate reader (Epoch 2 Microplate Spectrophotometer, BioTek, USA with Gen5 Microplate data connection and analyses software) was used to check and monitor growth by OD change. Parallel shake flask experiments were set up to define a correlation between OD measurements obtained from microplate reader (OD <sub>microplate</sub> ), UV-Vis spectrophotometer (OD <sub>UV-vis</sub> ) (Biochrom™ Biowave II) and dry biomass measured at 37°C. OD changes recorded and obtained correlations between OD measurements and dry biomass were used to calculate growth rate and growth yield from 27 to 45°C.
Data format	Raw and filtered
Description of data collection	Microplate experiments were conducted at selected temperature range (27 to 45°C) in 48 wells microplates for 24h to 1 week to monitor and record <i>E. coli</i> growth. <i>E. coli</i> growth in parallel shake flask experiments were monitored with UV-Vis spectrophotometer. Dry biomass was measured at the end of exponential growth phase of the culture with dry mass analysis.
Data source location	INRAE, Antony, Paris, France
Data accessibility	Repository name: Recherche Data Gouv Data identification number: UNF:6:vUb2nADFkt6NojFpCTI3Sw== [fileUNF] Direct URL to data: <a href="https://doi.org/10.57745/GCKG7W">https://doi.org/10.57745/GCKG7W</a>

Value of the Data

- These data are valuable for mechanistic studies requiring robust and accurate joint measurement of growth rates and yields at different temperatures, together with thermodynamic calculation of mass and energy balances of microbial growth. OD change, growth yield and growth rate data may be further used by modelers in mechanistic studies of microbial growth.

## 1. Objective

Mechanistic studies about the influence of temperature on microbial growth requires robust and accurate joint measurement of growth rates and yields at different temperatures in well-defined culture medium which chemical composition can be precisely specified in order to compute thermodynamic calculation of mass and energy balances of microbial growth. This data set reports influence of temperature on *E. coli* growth rate and yield both at the same time for minimal media where glucose was the sole carbon and energy source. Growth rates were measured in 28 to 40 parallel cultures using OD curves obtained with a thermostated microplate reader. Yields were obtained from the same OD curves using conversion factors estimated from dedicated experiments where suspended dry biomass measurements and UVvis spectrophotometer measurements were performed in parallel.

## 2. Data Description

*E. coli* was grown in modified MOPS medium with 0.5 g/L of glucose in 48 wells microplates at 18 different temperatures ranging from 27 to 45°C. OD measured for each well every 10 min in the microplates (OD<sub>microplate</sub>) are reported as “raw” in the file names [1]. On each microplate, 6 wells were used as negative control with modified MOPS medium without glucose. The accuracy of the instrument is +/- 0.010 OD unit in the 0 to 2 OD unit range.

### 2.1. Data filtration

The raw data for each well were manually checked and the wells for which OD values did not increase were considered as non-growing wells and were discarded. OD<sub>microplate</sub> for growing wells are reported as “filtered” in the file names. Average and standard deviations for growth rates and yields were computed for each temperature from these filtered data (see Table 1).

### 2.2. Growth rates

The exponential phase was determined through the use of a semi-log plot representation of OD<sub>microplate</sub> vs. time. For each growing well (see data filtration above), the slope in the linear portion of the curve was used for growth rate estimation. Average and standard deviation of growth rates of *E. coli* calculated for temperatures ranging from 27 to 45°C are shown in Fig. 1.

### 2.3. Growth yield

*E. coli* growth yield was calculated for each temperature and each growing well from the growth curves. The maximum and minimum OD<sub>microplate</sub> in each growth curve was first converted to OD<sub>UVvis</sub> and the change in optical density  $\Delta OD_{UV-vis}$  was calculated. The yield,  $Y_{XS}$ , was expressed as the ratio of the dry mass of *E. coli* to consumed glucose concentration during microbial growth. Dry biomass was obtained by using the conversion factor given in OD-dry biomass correlation section. Average yields and standard deviations calculated for the different temperatures are shown in Fig. 2.

$$Y_{XS} = \Delta OD_{UV-vis} * \frac{\text{OD} - \text{dry mass conversion factor}}{\text{glucose concentration}}$$

**Table 1**

Means and standard deviations of growth rates and yields calculated for temperatures ranging from 27 to 45°C. The number of wells considered for the calculations of means and standard deviations are indicated in the last column. Most of the calculations were performed using data from 40 wells, however in some cases growth was not observed in some wells and the corresponding data was discarded (see data filtration above).

Temperature °C	Yield C mol biomass/C mol glucose	$\mu$ 1/h	Number of wells with growing culture
27	0.71±0.02	-	28
28	0.69±0.03	0.65±0.02	32
29	0.71±0.02	0.71±0.01	40
30	0.68±0.04	0.72±0.02	40
31	0.70±0.01	0.80±0.01	40
32	0.69±0.02	0.86±0.02	40
33	0.66±0.02	0.92±0.02	39
34	0.67±0.01	0.94±0.02	40
35	0.65±0.02	1.01±0.02	39
36	0.63±0.02	1.03±0.03	40
37	0.62±0.02	1.03±0.06	40
38	0.61±0.03	1.04±0.07	39
39	0.59±0.04	1.07±0.06	40
40	0.58±0.03	1.06±0.07	40
41	0.52±0.03	1.07±0.09	35
42	0.55±0.02	1.13±0.06	40
43	0.56±0.03	1.02±0.04	37
44	0.53±0.03	1.01±0.07	39
45	0.35±0.05	0.72±0.14	40

2.4. Estimation of conversion factor of OD<sub>UV-vis</sub> to dry biomass

*E. coli* culture growth in modified MOPS media with 0.5 g/L of glucose was monitored and recorded with UV-vis spectrophotometry for 7.2 h at 37°C. At the end of the culture, dry mass of grown cells and the amount of glucose consumed were measured for duplicate samples where standard deviation was 1,6%.

Table 2 shows the conversion from OD<sub>UV-vis</sub> to dry biomass calculated from this experiment. It is important to note that maximum value of OD<sub>UV-vis</sub> readings was 0,85 OD in culture experiments. However, the correlation was normalized to 1 OD to be able to compare with literature.

**Table 2**

Conversion factor between *E.coli*, dry mass, and OD<sub>UV-vis</sub> readings.

	OD <sub>UV-vis</sub> , at 600 nm	dry weight, g/L	g dry weight/OD <sub>UV-vis</sub> /ml
This study	1	0.424	4.2E-04
Myers et al. [3]	1	0.396	3.96E-04

2.5. Conversion of OD<sub>Microplate</sub> to OD<sub>UV-vis</sub> for *E. coli*

OD of serial dilutions of *E. coli* culture grown at 37°C in modified MOPS medium with 3 g/L of glucose were measured with UV-vis spectrophotometry and microplate reader to establish a calibration curve. The values of OD<sub>UV-vis</sub> as function OD<sub>Microplate</sub> is shown in Fig. 3 and the related data is available [here](#).

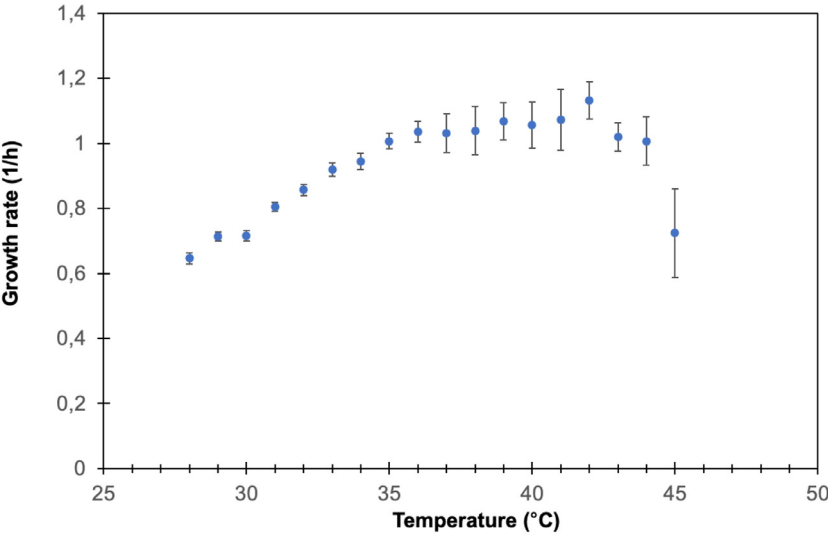


Fig. 1. Means and standard deviations of growth rates of *E. coli* as a function of temperature.

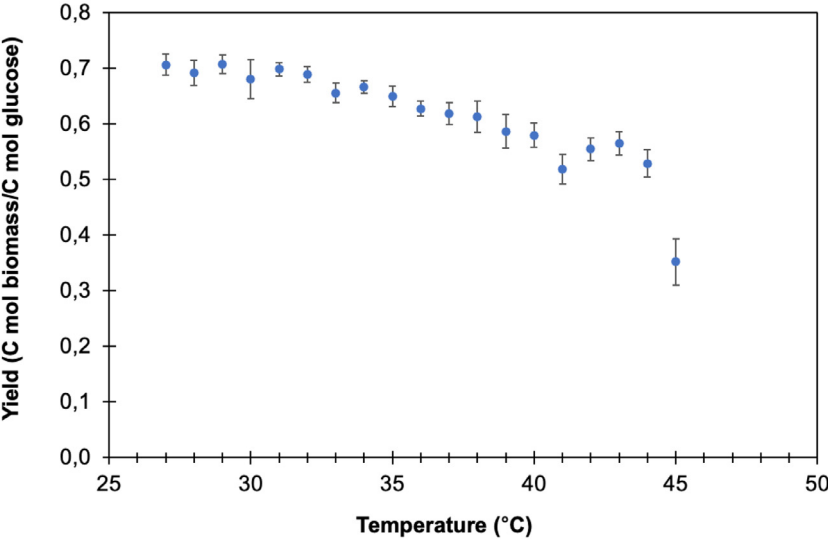
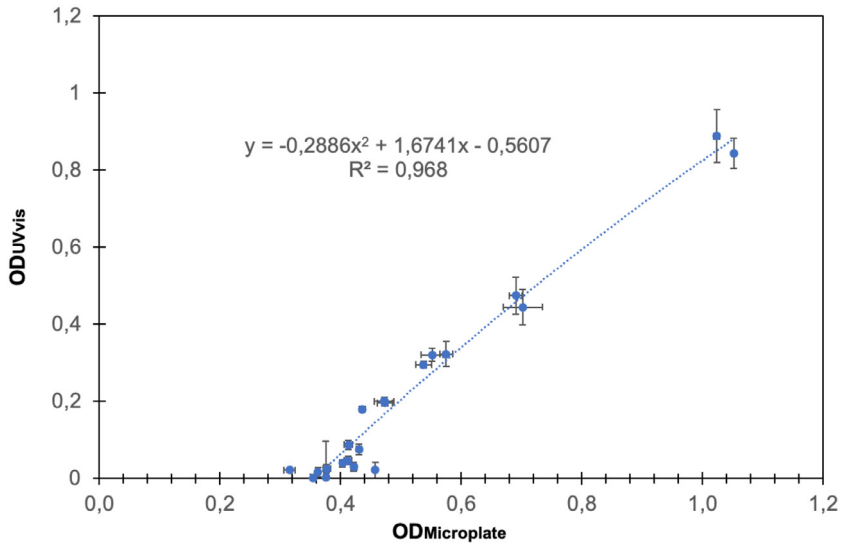


Fig. 2. Means and standard deviations of growth yields of *E. coli* as a function of temperature.



**Fig. 3.** Means and standard deviations  $OD_{UV-vis}$  vs. means and standard deviations of  $OD_{microplate}$ . The formula of a second order polynomial regression and the associated  $R^2$  value are shown on the graph.

### 3. Experimental Design, Materials and Methods

#### 3.1. Microbial Culture

*Escherichia coli* K12 NCM3722 strain ( $\Delta motA$ ), was selected as test organism [2]. This non-motile *E. coli* K12 NCM3722 strain ( $\Delta motA$ ) was obtained from Taheri Lab (California State University, 18111 Nordhoff St., Northridge, CA 91330).  $-80^\circ\text{C}$  frozen pure culture aliquots were used to inoculate modified MOPS medium with 0.5 g/L glucose in order to obtain precultures in microplate, standardization and correlation experiments. Precultures and cultures were grown at the same target temperature (from 27 to  $45^\circ\text{C}$ ). MOPS medium was prepared with the following composition: (40 mM), tricine (4.0 mM), iron sulfate stock (0.01 mM), ammonium chloride (9.5 mM), potassium sulfate (0.276 mM), calcium chloride (0.0005 mM), magnesium chloride (0.525 mM), sodium chloride (50 mM), ammonium molybdate ( $3 \times 10^{-9}$  M), boric acid ( $4 \times 10^{-7}$  M), cobalt chloride ( $3 \times 10^{-8}$  M), cupric sulfate ( $10^{-8}$  M), manganese chloride ( $8 \times 10^{-8}$  M), zinc sulfate ( $10^{-8}$  M), potassium phosphate dibasic (1.32 mM) [2]. Modified MOPS medium was obtained by adding 5  $\mu\text{g/ml}$  of following amino acids: L-methionine, L-histidine, L-arginine, L-proline, L-threonine, and L-tryptophan.

#### 3.2. Microplate Experiments

A microplate Reader (Epoch 2 Microplate Spectrophotometer, BioTek, USA) was used to check and monitor microorganisms' growth by OD change. Gen5 Microplate data connection and analyses software was used for the analyses of microplate data. Microplate experiments were performed at different temperatures ( $27-45^\circ\text{C}$ ) in 48 well microplates for 24h to 1 week. The plates were shaken with a double orbital shaking pattern with 807 cycles per minute with an amplitude size of 1 mm.  $1^\circ\text{C}$  temperature gradient was applied to minimize evaporation on the top of microplate lid and  $OD_{600}$  corrected for pathlength was monitored for each well.

### 3.3. OD Measurement Standardization and OD-Dry Biomass Correlation

A modified version of a method proposed by Myers et al. [3] was applied for standardization and calibration of OD results from microplate readings.

28  $\mu\text{L}$  of overnight preculture was used for inoculation of 56 mL of cultivation medium in 250 mL erlen flasks (dilution factor  $5.10^{-4}$ ). *E. coli* cultures were grown in modified MOPS medium containing 3 g/L glucose at  $37^{\circ}\text{C}$  until reaching stationary phase. A relatively high glucose concentration was used to test high number of serial dilutions. In order to standardize microplate readings, both UV-Vis spectrophotometer and microplate reader was used to determine cell densities of grown culture for a series of dilutions.

Triplicate MOPS *E. coli* culture were diluted in series. Cultures were mixed with PBS solution to have from 0.001 to 0.5 times dilutions in a total volume of 11.1 mL. 21 diluted cultures were measured in parallel in UV-Vis spectrophotometer and microplate reader. PBS was used as blank in UV-Vis spectrophotometer whereas  $\text{OD}_{\text{Microplate}}$  measurements were not blanked. Data points were used to derive polynomial relationship between UV-Vis and microplate optical densities.

The relationship between cell density and dry biomass is required for proper estimation of growth yield and other kinetic parameters. For this purpose, overnight *E. coli* precultures were inoculated in 2 L erlen flasks with modified MOPS medium containing 0.5 g/L glucose at  $37^{\circ}\text{C}$ . 225  $\mu\text{L}$  of overnight preculture was used for inoculation of 450 mL medium (dilution factor  $5.10^{-4}$ ) and bacterial culture growth was monitored with  $\text{OD}_{\text{UVVis}}$  readings until stationary phase for around 7 hours. At 7.2h the cultures were directly used for dry mass analysis as defined in standard methods [4]: 40 mL bacterial culture was filtered through 0.22  $\mu\text{m}$  filters. Filters were dried at  $105^{\circ}\text{C}$  for 24h and transferred to desiccator until stabilized filter weights were measured. Dry biomass corresponding to 1 OD was calculated (0.42 mg/OD/mL). This value is close to the  $0.396 \pm 0.011$  mg/OD/mL value reported for *E. coli* (DH 5 $\alpha$ ) in literature [3].

### 3.4. Glucose Measurements

Glucose measurements were performed with Colorimetric Glucose Assay kit (CellBioLabs). Samples were filtered before analysis using 0.22  $\mu\text{m}$  filters. Samples and glucose standards were incubated for 30–45 minutes in 96-well microtiter plates and read with colorimetric plate reader (Epoch 2 Microplate Spectrophotometer, BioTek, USA). Glucose concentration in samples was determined by comparing the microplate readings with readings for glucose standards with known concentrations.

## Ethics Statements

This work does not include any studies with human or animal subjects.

## CRediT Author Statement

**Tugce Katipoglu-Yazan:** Conceptualization, data acquisition, writing-review and editing **Subrata Dev:** Conceptualization, data acquisition, writing-review and editing **Elie Desmond-Le Quémener:** Conceptualization, writing-review, editing and supervision **Théodore Bouchez:** Conceptualization, funding acquisition, writing-review, editing and supervision.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the studies in this paper.



## Data Availability

Data on the influence of temperature on the growth of *Escherichia coli* in a minimal medium containing glucose as the sole carbon source for the joint computation of growth yields and rates at each temperature from 27 to 45°C (Original data). (Recherche Data Gouv).

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