“Problems worthy of attack prove their worth by hitting back.”

—Piet Hein

1. **Snapshot—evolution by the numbers.**
Read the two versions of snapshot that are attached with this homework. Your job in this homework is to make a version of your thinking on Evolution by the Numbers in which you have as many as 5 categories (see the Snapshot on Cell Biology where the categories are concentrations, sizes, rates, etc) and can have as many as 5 numbers per category. In addition, please also examine the supplemental material for the snapshot that gives information on the data leading to each of the different numbers. Please produce such a supplement of your own that gives the sources for your own numbers. Please submit your answer to Rob, Justin, Bill, and Manuel in PDF form by email.
### Characteristic rates and timescales in cell biology

**Model bacterium (E. coli)** vs. **Mammalian cell line (HeLa)**

**Diffusion over 1 μm**
- 10-100 ms

**DNA replication**
- 10⁷ nt/s

**Channel**
- 0.1 μs

**Transporter**
- 1-10 ms

**Passage across membrane**
- 1 min

**DNA replication**
- 10³ nt/min

**Cell movement**
- 10 μm/s vs. 1 μm/min

**Cell cycle**
- 1 hr vs. 1 day

**Flagellar rotation**
- 100 Hz

**Transcription**
- 10-100 nt/s
  - 1 min/gene [1 kbp]
  - 10 min/gene [10 kbp]

**Translation**
- 10 aa/s
  - 1 min/protein [300 aa]

**Cellular pool half-life** (dominated by)
- 1 s: Metabolite (turnover)
- 10 min: mRNA (degradation)
- 1 hr: Protein (dilution)

**Orders of magnitude in timescales**

<table>
<thead>
<tr>
<th>Fastest enzyme turnover time</th>
<th>Neuronal coincidence detection</th>
<th>ATP synthase rotation</th>
<th>Protein folding</th>
<th>Gene splicing</th>
<th>Budding yeast generation time</th>
<th>Taste bud cell lifespan</th>
</tr>
</thead>
<tbody>
<tr>
<td>10⁻⁶ (μs)</td>
<td>10⁻³ (ms)</td>
<td>10⁰ (s)</td>
<td>10³ (=20 min)</td>
<td>10⁶ (=2 weeks)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- Electron transfer by cytochrome c
- Action potential duration
- Average enzyme turnover time
- Protein translation
- Minimal bacterial generation time
- Circadian clock
- Red blood cell lifespan

Sizes not to scale
V_{bacteria} ∼ V_{cell line} ~1 : 1,000

**See online version for legend and references.**
SnapShot: Timescales in Cell Biology

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Knowing key timescales enables us to quickly gain intuition, perform sanity checks and serves as a sixth sense in understanding how cells grow and communicate. This SnapShot briefly communicates some of the insights we collected about rates and durations of these processes from thinking about cell biology by the numbers.

Can Metabolism Wait for Gene Expression?

Metabolic networks and gene regulatory networks, the epicenters of biological regulation, exert their control on distinct timescales. The characteristic concentration of a metabolite in central metabolism is on the order of 1 mM, while the flux in the cellular metabolic highway of glycolysis is usually on the order of 1 mM/s in bacteria and 0.1–0.01 mM/s in mammalian cell lines. Thus, the turnover time is on the order of a second for bacteria and a minute for mammalian cell lines. If the production and consumption reactions are not in balance, the metabolite pool will be consumed (or compounded) rapidly, before gene expression of metabolic enzymes—which takes minutes—can do anything about it. Instead, allosteric regulation and post-translational modification are used to control metabolic flux.

How Long Does It Take to Get a Functional GFP?

Induction of GFP expression begins with the addition of an inducer to the medium. The inducer diffuses into the cell or binds to a receptor to activate transcription within seconds. Transcription and translation in bacteria take on the order of a minute, with protein folding occurring concurrently. However, a maturation process, which involves cyclization and oxidation of the GFP chromophore, takes tens of minutes. Evolved versions reduce the maturation time so that the whole process, from induction to fluorescence, is achieved within minutes.

Which Processes Govern the Half-Life of Cellular Components?

Cellular components, such as metabolites, mRNA, and proteins, have different timescales for turnover. The cellular half-life of a metabolite results from the ratio between the metabolite pool size and the flux through it. For mRNA, the half-life is dictated by nuclease degradation. Proteins are much more stable and are usually degraded at timescales longer than the fast doubling time achievable in the lab, which makes dilution by cellular division the predominant process controlling their turnover. These are, of course, broad generalizations and biology thrives on bending them; for example, rapid regulated proteolysis leads to much faster protein turnover.

Are Mammalian Cells Slow-Motion Versions of Bacterial Cells?

Bacteria and mammalian cells work under similar physical and chemical constraints. For example, the diffusion coefficients and the rates of the RNA polymerase and ribosomes are similar. Yet, with a larger cell size and gene length, the functional timescales in a mammalian cell are extended. For example, diffusion of a protein across a cell will take ~0.1 s in a 1 µm bacterial cell and ~10 s in a 10 µm mammalian cell. Similarly, an average bacterial gene is 1 kbp long and thus will take about a minute to transcribe, while introns cause the average mammalian gene to be 10 kbp long and thus will take about 10 min. Similar timescale differences occur in additional cellular processes, such as the turnover of metabolites. For such key processes, what is true for a bacterium on a 1 second timescale is true for a mammalian cell in about 1 min.

How to Get a Protein across a Neuron on Time?

For a protein to get from the tip of the axon to the soma in a 1 cm long neuron, two main mechanisms are possible. Diffusion (D = 10 µm²/s) would take over a month (scaling like R²/D). Alternatively, a molecular motor with a speed of 1 µm/s can transport the protein within a few hours. In neurons over a meter long—in humans or giraffes for example—even with molecular motors, the journey should take several days.

A Speed Limit on Crawling Cells?

Cellular motility is powered by actin polymerization at the lamellipodium leading edge. The speed limit for a growing actin network is the growth rate of a single filament oriented perpendicular to the leading edge. The on-rate for the addition of an actin monomer to the growing tip is 10⁻¹⁰ M⁻¹ s⁻¹. The reported cellular concentration of polymerizable actin monomers ranges between 1 and 100 µM, and in such cases, we choose to use the geometric mean (10 µM). Each polymerized actin monomer adds 3 nm to the filament, and we thus get a velocity on the order of 1 µm/s (3 × 10⁻¹⁰ M⁻¹ s⁻¹ × 10 µM × 3 nm), which is observed, for example, for Listeria. The observed crawling speeds of fish keratocytes and mammalian fibroblasts are one and two orders of magnitude slower, respectively. The counteracting membrane tension, cell adhesion, and the fact that a lamellipodium is an ensemble of filaments reduce the speed of cells below the polymerization-based speed limit.

What is the Lifespan of Different Cells in Our Body?

The intestine epithelium turns over in less than a week, our skin epidermis in a week to a month, and if you burn your tongue, taste buds return in about 2 weeks. Red blood cells have a lifespan of 4 months, such that donating 0.5 L from our 5 L of blood every few months does not deplete them. A striking difference in lifespan exists between sperm cells (~50 days) and oocytes (~50 years). Fat cells and skeleton replace themselves in about 10 years, while most of the meso-in the central nervous system and our eye lens cells are not replaced at all throughout our life.

How Fast Can Olympic Athletes Respond to the Starter’s Pistol?

Upon hearing the shot, athletes process and propagate an electric impulse from the brain all the way to their feet (~1 m). Considering the speed of the action potential (10–100 m/s), this implies a latency of 10–100 ms regardless of other processes, such as the speed of sound and signal processing in the brain. The best athletes respond after ~120 ms, and a reaction time below 100 ms is immediately disqualified as a false start.

ACKNOWLEDGMENTS


REFERENCES

Cell size

- **Bacteria (E. coli):** 0.7-1.4 μm diameter, ~2-4 μm length, ~0.5-5 μm² in volume; 4×10⁻¹⁵ cell/ml for culture with OD₆₀₀≈1
- **Yeast (S. cerevisiae):** ~3-6 μm diameter, ~20-160 μm³ in volume
- **Mammalian cell volume:** 100-10,000 μm³; HeLa cell: 500-5000 μm³ (adhering to slide ~15-30 μm diameter)

Length scales inside cells

- **Nucleus volume:** ~10% of cell volume
- **Cell membrane thickness:** ~4-10 nm
- **“Average” protein diameter:** ~3-6 nm
- **Base pair:** 2 nm (D) x 0.34 nm (H)
- **Water molecule diameter:** ~0.3 nm

Energetics

- **Membrane potential:** ~70-200 mV → 2-6 k_BT per electron (k_BT = thermal energy)
- **Free energy (ΔG) of ATP hydrolysis under physiological conditions:** ~40-60 kJ/mol → ~20 k_BT/molecule ATP; ATP molecules required during an E. coli cell cycle ~10⁻¹¹ 10⁻¹²

Diffusion and catalysis rate

- **Diffusion coefficient for an “average” protein:** in cytoplasm D~5-15 μm²/s → 10 ms to traverse an E. coli; → ~10 s to traverse a mammalian HeLa cell; small metabolite in water D~500 μm²/s
- **Diffusion-limited on-rate for a protein:** ~10⁻⁸⁻¹⁰⁻¹⁵ M⁻¹s⁻¹ → for a protein substrate of concentration ~1 μM the diffusion-limited on-rate is ~100-1000 s⁻¹ thus limiting the catalytic rate k_cat

Concentration

- **Concentration of 1 nM:** in E. coli ~1 molecule/cell; in HeLa cells =1000 molecules/cell
- **Characteristic concentration for a signaling protein:** ~10 nM-1 μM
- **Water content:** ~70% by mass; general elemental composition (dry weight) of E. coli: C6H12O6N4, Yeast: C6H10O5N4
- **Composition of E. coli (dry weight):** ~55% protein, 20% RNA, 10% lipids, 15% others
- **Protein concentration:** ~100 mg/ml = 3 mM; 10⁻¹⁰⁻¹⁰⁻⁰£P (E. coli (depending on growth rate)); Total metabolites (MW < 1 kDa) ~300 mM
- **Membrane potential:** per electron: E. coli = 20-40 min; budding yeast 70-140 min; HeLa human cell line: 15-30 hr
- **Rate of replication by DNA polymerase:** E. coli = 200-1000 bases/s; human ~40 bases/s. Transcription by RNA polymerase 10-100 bases/s
- **Translation rate by ribosome:** 10-20 aa/s
- **Degradation rates (proliferating cells):** mRNA half life < cell cycle time; protein half life ~cell cycle time

Diffusion, replication, transcription, translation, and degradation rates

- **Cell cycle time (exponential growth in rich media):** E. coli = 20-40 min; budding yeast 70-140 min; HeLa human cell line: 15-30 hr
- **Generation time:** E. coli = 20-40 min; budding yeast ~200 min; HeLa human cell line: 15-30 hr

Genome sizes and error rates

- **Genome size:** E. coli ~5 Mbp
- **Number of protein-coding genes:** E. coli ~4000; S. cerevisiae ~6000; C. elegans, A. thaliana, M. musculus ~15,000
- **Mutation rate in DNA replication:** ~10⁻⁶⁻¹⁰⁻¹⁰ per bp

Useful biological numbers extracted from the literature. Numbers and ranges should only serve as “rule of thumb” values. References are in the online annotated version at [www.BioNumbers.org](http://www.BioNumbers.org). See the website and original references to learn about the details of the system under study including growth conditions, method of measurement, etc.

See online version for legend and references.
SnapShot: Key Numbers in Biology

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Biology is becoming increasingly quantitative. Taking stock of key numbers in cell and molecular biology enables back-of-the-envelope calculations that test and sharpen our understanding of cellular processes. Further, such calculations provide a quantitative context for the torrent of data from new experimental techniques. However, such useful numbers are scattered in the vast biological literature in a way that often leads to a frustrating literature-mining ordeal. Here, we have collected a set of basic numbers in biology that we find extremely useful for obtaining an order of magnitude feel for the molecular processes in cells. Several examples (see below) show how to combine these numbers to think about biological questions. The values should be considered rules of thumb rather than definitive values as variety is the spice of life and variability is ever present in biology. This compilation is based on the BioNumbers wiki project (http://www.BioNumbers.org) where these and the values of several thousand other biological properties are provided together with their experimental context and references to the primary literature.

Is There Enough Time to Replicate the Genome?
The bacterium Escherichia coli has a genome of roughly 5 million base pairs (bp) and a replication rate in the range of 200–1000 bp/s. These numbers imply that it should take two replisomes at least 2500 s to replicate the genome, a number that is much larger than the maximal division rate of ~20 min. How can this be? It turns out that, under ideal conditions, E. coli uses nested replication forks that begin to replicate the DNA for the granddaughter and great granddaughter cells before the daughter cells have even completed replication.

How Many Mutations in a 5 ml Culture of Bacteria?
Using the 10⁻¹⁰ bp mutation rate of E. coli per replication and a genome size of ~10⁹ (both strands), we predict ~10⁻⁴ mutations per genome replication. In a 5 ml saturated culture (optical density ~2.0) of E. coli, there are about 10⁶ to 10⁹ cells. The final doubling of this culture requires the replication of ~10⁶ cells; thus even this last cell division event would be responsible for ~10⁸ single base pair substitutions. If the culture started with a single bacterium, every single nonletal base pair substitution in the E. coli genome is likely to be represented in the culture.

How Long to Reach Confluence?
In a 96 multiwell plate, each well has a diameter of 5 mm (i.e., an area of ~20 mm² = 2 x 10⁻² m²). Given that the diameter of a HeLa cell is ~25 µm (i.e., ~500 µm² area), it takes roughly 40,000 cells to reach confluence. Starting with a single cell (obtained by cell sorting rather than cell splitting) with a generation time of about 1 day, the time to reach confluence is about 2 weeks.

How “Dense” Is a Saturated E. coli Culture?
A saturated E. coli culture has about 10⁹ cells/ml. Given that each cell is about 10⁻¹⁵ grams, we get a cell concentration of about 1 mg/ml or about 1 part in a thousand of the mass (or volume). The mean spacing between the cells is roughly 10 µm (which is not as dense as the concentration of bacteria in the gut of the termite where densities are typically a factor of ten higher).

How Many Carbon Atoms Are in a Cell?
A cell with a volume of 1 µm³ and a density of about 1 g/ml has a total mass of 10⁻¹⁲ grams. From the formula C₆H₁₂O₆N and the weights of the elements, we derive a carbon content of about 12 x 4/(12 + 4 + 7 + 2 x 16 + 14) = 48/101 or about one half of the dry mass. With 30% dry mass (70% water), we obtain ~10⁻¹³ gm of carbon. Next we transformed the number of molecules using Avogadro’s constant: 6 x 10⁶⁰ x 10⁻¹³/12 = 5 x 10⁴ carbon atoms per cell. To verify this, we have done the calculation in a different way; assuming there are about 3 x 10⁹ proteins, each one consisting of about 300 amino acids, we get a total of ~10¹⁰ amino acids. An amino acid has about five carbon atoms, so we arrive at a similar value. Both estimates depend linearly on the cell volume, which can vary significantly based on growth conditions.

How Far Can Macromolecules Move by Diffusion?
It takes about 10 s on average for a protein to traverse a HeLa cell. An axon 1 mm long is about 100 times longer than a HeLa cell, and as the diffusion time scales as the square of the distance it would take 1⁰⁶ seconds or ~2 days for a molecule to travel this distance by diffusion. This demonstrates the necessity of mechanisms other than diffusion for moving molecules long distances. A molecular motor moving at a rate of ~1 µm/s will take a “reasonable” time (~15 min) to traverse an axon 1 mm in length.

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REFERENCES
**Supplementary information – SnapShot: Basic numbers for cell biology:**

**Cell Sizes:**

1. **Bacteria (E. coli):** ~1 µm diameter (BNID 100002)
   
   **Ref:** Nelson DE, Young KD. Penicillin binding protein 5 affects cell diameter, contour, and morphology of Escherichia coli. J Bacteriol. 2000 PMID 10692378

   **Measurement Method:** Cell measurements and analyses were performed with Image Pro. Cell diameters of individual cells were calculated by measuring the cross-sectional area and dividing that value by the cell length.

   **Comments:** The derived diameters of 92% of untreated wild-type E. coli CS109 cells clustered between 1.0 and 1.1 µm (Fig. 3A)

   ~2.5 µm length (BNID 103713)


   **Measurement Method:** Electron micrographs were projected at a final magnification of about 15,000x to 20,000x, and cell length and cell diameter were measured. At least 150 cells were measured for each sample, and dimensions were calculated according to a calibration grid which was photographed at the same magnification.

   **Comments:** Value is mean of E. coli B IlvA, thyA, cells grown on glycerol and glucose during exponential phase, Mean doubling time 57 min.

   ~1.1 µm³ in volume 0.44-1.79 mm³ (BNID 100004)


   **Measurement Method:** Coulter Counter analyzer system

   **Comments:** value is average of 10 measurements in table 1

   2×10^7 cell concentration for culture with OD600 of 0.1 (BNID 103625)

   **Ref:** Sezonov G, Joseleau-Petit D, D’Ari R. Escherichia coli physiology in Luria-Bertani broth. J Bacteriol. 2007 Fig. 1a PMID 17905994

   **Measurement Method:** The widely used wild-type E. coli K-12 strain MG1655 was chosen for this study. An overnight culture in Luria-Bertani broth was diluted 5,000-fold in 250 ml fresh medium in a 1-liter Erlenmeyer flask and cultivated in a shaking water bath (180 rpm) at 37°C. The OD600 and number of cells per milliliter were monitored.

   **Comments:** Note that cell concentration of a different strain with OD600 of 0.1 is 10³⁸ cells/ml, greater by a factor of 5 (see BNID 100985). The optical density of yeast and bacterial cultures is due to light scattering, not absorption. Therefore it is sensitive to the geometry of the detector relative to the cuvette, as well as the size and shape of the cells.

2. **Yeast (Saccharomyces cerevisiae):** ~5 µm diameter (BNID 103896)


   **Measurement Method:** The cell diameters were measured using a Coulter counter and an image analyser. Micromanipulation was used for measuring the bursting strength of individual yeast cells. The basic principle of this technique is the capture and squeezing of a single cell between parallel surfaces.

   **Comments:** S. cerevisiae Yg grown in YEPG medium at 60% DO₂.

   ~70 mm³ in volume (BNID 100452)

   **Ref:** Roskams and Rodgers, LabRef

3. **Volume of mammalian (HELA) cell 4400-5000 µm³ (BNID 103719)**


   **Measurement Method:** Cells allowed to enter logarithmic phase. Cell number was obtained with a Coulter counter, while cell sizing was performed with the automatic particle size distribution analyser Model J Electronic Co., Hialeah, Fla.

   **Comments:** Please note-BNID 103725 gives HeLa cell volume range of 760-2730 um³
Human erythrocyte 97 µm³ in volume (BNID 101711)


COS-7 cell 2016±209 µm³ (BNID 101666)

Ref: Bohil et al. Myosin-X is a molecular motor that functions in filopodia formation. PNAS 2006. 103(33): pp.12411-12416 PMID 16894163

Comments:value is found in supplemental figure 7. The COS cell line was obtained by immortalizing a CV-1[1] cell line derived from kidney cells of the African green monkey.

Human alveolar macrophage cell 4990±174 µm³ (BNID 103566)


Measurement Method: Morphometric analysis of AM was performed using a flow cytomter that generates volume signals based on the Coulter-type measurement of electrical resistance.

Perikaryon of Purkinje cell in human cerebellum 13700 µm³ (BNID 103180)


Measurement Method: stereological tools designed for estimating mean particle volume: The nucleator and the rotator.

Comments:Cerebella from 19 normal Caucasian males, ages 19-84 years, were studied. The perikaryon is also known as the cell body.

Organelles and cell constituents:

4. Fraction of Budding yeast nucleus volume out of total cell volume ~7% (BNID 104708)

Ref: Jorgensen P, Edgington NP, Schneider BL, Rupes I, Tyers M, Futcher B. The size of the nucleus increases as yeast cells grow. Mol Biol Cell. 2007 Sep18(9):3523-32. Table 1 Table link – http://tinyurl.com/ncgbno PMID 17596521

Measurement Method: Morphometry

Comments: Analysis of mutant yeast strains spanning a range of cell sizes revealed that the ratio of average nuclear volume to average cell volume was quite consistent, with nuclear volume being approximately 7% that of cell volume. See Nuclear volume of 2.9 um^3 BNID 100447

HeLa cell nucleus in cervix tissue ~400 µm³ in volume (BNID 101402)

Ref: Maul GG, Deaven L. Quantitative determination of nuclear pore complexes in cycling cells with differing DNA content. J Cell Biol. 1977 Jun73(3):748-60.3. Table 1 Table link – http://tinyurl.com/58x8ay PMID 406262

Measurement Method: HeLa, mouse L, and CHO cells were grown in suspension culture. Their nuclei appeared mostly round. The long and short axes of these nuclei were averaged, and the surface and volume were calculated as if the nuclei were spheres.

5. Cell membrane thickness ~4-10 nm

(BNID 103944) 5-10 nm for E. coli


(BNID 101276) 4.1 nm for spinach


Measurement Method: atomic force microscopy

(BNID 103948) 7.5 nm for vertebrate erythrocyte

6. "Average" protein diameter ~4 nm (BNID 101827)
Page 26
Comments: See Stokes radius of Casein 2.9-3.7 nm (BNID 103862), Radius of gyration of myoglobin 1.52 nm (BNID 103782), Radius of gyration of ribonuclease 1.43 nm (BNID 103781), Radius of gyration of conalbumin 2.95 nm (BNID 103737), albumin diameter 3 nm (BNID 103736), insulin receptor diameter 6.5 nm (BNID 103212), Diameter of the LacY transporter ~6 nm (BNID 102929)

Properties of casein, BSA, and lysozyme Table link - http://tinyurl.com/cu28na (BNID 103880)
Ref: Pim van Hee, Selective recovery of micrometer bioparticles using aggregation and air flotation, thesis, 2006, Department of Biotechnology, Delft University of Technology The Netherlands
Comments: Radii of casein, BSA, and lysozyme 2.9-3.7, 3.48, and 1.8 nm, respectively
Radius of gyration of ribonuclease 1.43 nm (BNID 103781)
Measurement Method: X ray crystallography
Comments: Estimated hydrated protein radius is 1.71 nm

Radius of gyration of myoglobin 1.52 nm (BNID 103782)
Measurement Method: X ray crystallography
Comments: Estimated hydrated protein radius is 1.84 nm

7. Water molecule diameter ~0.28 nm (BNID 103723)
Comments: Other sources give value of 2.63 Angstrom

Concentrations:
8. Concentration of 1 nM in a cell the volume of E. coli is ~ 1 molecule/cell (BNID 102068)
Ref: Calculated according to cell volume in Primary Source, please see Measurement Method
Measurement Method: Calculated using Avogadro’s constant according to one cell volume=1E-12 cm^3=1E-15 liter (Average of 10 measurements in table from Primary source, BNID 100004). Calculated using Avogadro’s constant according to one cell volume=1E-12 cm^3=1E-15 liter. 1 particle/1E-15 liter=X particles/liter >> X=1E15 particles/liter 1E15(particles/liter)/6E23(particles/mole)=1.666E-9M=1 nM.
Concentration of ~0.1 pM in a mammalian (HeLa) cell is ~ 1 molecule/cell (BNID 104519)
Ref: Calculated according to cell volume, please see Measurement Method
Measurement Method: Calculated using Avogadro’s constant according to one cell volume=5000 um^3=5E-12 liter (BNID 103719). 1 particle / 5E-12 liter = X particles/liter >> X = 2E11 particles/liter 2E11 (particles/liter)/6E23 (particles/mole) = ~3E-13 M=0.1 pM. Thus 1 nM=10^{10} molecules per cell. Note: 3E-13 M and 1E-13 M are on same order of magnitude, allowing approximation.

9. Characteristic concentration for a signaling protein ~10 nM-1 mM (BNID 103954)
4:190 Table link - http://tinyurl.com/5sm66y PMID 18463614

**Primary Source:** See table for primary sources

**Measurement Method:** The Table gives a characteristic range of signaling proteins 10000-1000000 molecules per cell. Calculated using Avogadro's constant according to one cell volume (mammalian cell culture volume BNID 100434) ~2E-15cm^3 =2E-12 liter. 2E-12X6E23=~1E12 particles per cell with concentration of 1 Molar. Concentration of 0.1-1 uM would give 1e4-1e6 molecules per cell.

**Comments:** Table gives Number of various signaling protein molecules in one cell. This table can be found in the supplementary information section of the above-mentioned reference. 0.4 mM concentration of Ras in HeLa cells (BNID 100853)

**Ref:** Fujioka et al., Dynamics of the Ras/ERK MAPK cascade as monitored by fluorescent probes. J Biol Chem. 2006 Mar 31281(13):8917-26 PMID 16418172

**Measurement Method:** FRET (Fluorescence Resonance Energy Transfer) probe

10. Water content: ~70% of cell

**E. coli** 74%, (BNID 100044)

**Ref:** Jerry W. King, Gary R, Supercritical fluid technology in oil and lipid chemistry, 1996, American Oil Chemists’ Society, pp. 303

**Yeast,** 68% (BNID 103689)

**Ref:** Jerry W. King, Gary R, Supercritical fluid technology in oil and lipid chemistry, 1996, American Oil Chemists’ Society, pp. 303

**Rat Alveolar macrophage** 67% (BNID 103960)


**Measurement Method:** Rat alveolar type I cells were isolated by enzymatic digestion and purified by centrifugal elutriation and specific surface adsorption. The identity of the harvested cells was confirmed using electronic cell sizing and transmission electron microscopy. Calculated according to cell volume of 1456 um^3=1.456e-12 liter and water content of 0.97 ul/10^6 cells=0.97*10^-12 Liter per cell

Empirical elemental formula for biomass of **E. coli** C:H_{1.77}:O_{0.49}:N_{0.24} (BNID 101800)


**Comments:** This means that for every mole of carbon, there is 1.77 moles of hydrogen, 0.49 moles of oxygen and 0.24 moles of nitrogen. From this the carbon content of a cell is about 48% of the dry weight

11. Composition of dry weight of an **E. coli:** ~55% protein, 20% RNA, 10% Lipids, 15% others (BNID 101436)


**Comments:** General information on quantities of macromolecules per cell

Composition of dry weight of an **S. cerevisiae:** protein 40% fiber 30% RNA 10% ashes 4% lipids 1% others 15% (BNID 104157)


**Measurement Method:** Water content, ashes, and crude protein (N X5.8) were determined according to the AOAC (ref 15) procedures. Total lipids were determined according to the Bligh and Dyer method (16). Soluble and insoluble fibers were quantified by treating the sample first with proteolytic enzymes (pepsin/pancreatin) to digest sample protein, followed by filtration to retain the insoluble fiber and precipitation of the soluble fiber from the filtrate with ethanol.

12. Number of proteins in an **E. coli** cell 2.35×10^6 (depending on growth rate) (BNID 102990)

Measurement Method: Calculated for an average cell of E. coli B/r in balanced growth at 37 degrees celsius in aerobic glucose minimal medium with a 40 minute mass doubling time. This cell is 44% through its division cycle.

Calculated concentration of total protein in E. coli cell ~4 mM (BNID 104726)

Measurement Method: Calculated for an average cell of E. coli B/r in balanced growth at 37 degrees celsius in aerobic glucose minimal medium with a 40 minute mass doubling time. This cell is 44% through its division cycle. Calculated from Protein number of 2.35e6 (See BNID 102990 and table link in Primary Source) and assuming cell volume of ~1um^3=1e-15liter (BNID 10004) and avogadro's constant=6e23 particles/mole (BNID 101907). (2.35e6proteins/cell)/[(6e23proteins/mole)*(1e-15liter/cell)]=~4e-3M

Total observed intracellular metabolite pool in E. coli 300 mM (BNID 104678)

Measurement Method: Researchers quantified metabolites by Liquid chromatography-mass spectrometry (LC-MS/MS) using an isotope ratio-based approach (8). As isotope-labeled standards for many metabolites are not available, they used uniformly 13C-labeled glucose medium to label the intracellular metabolome of E. coli (25). This enabled the use of commercially available unlabeled compounds as internal standards.

Comments: The total observed intracellular metabolite pool was approximately 300 mM. A small number of metabolites dominate the metabolome on a molar basis, with glutamate being the most abundant.

**Energetics:**

13. **DG** needed to achieve order of magnitude ratio of concentrations: ~6 kJ/mole
   -5.7 kJ/mole (BNID 103912)


Comments: Approximately 2kT=Approximately 60meV. Calculating RT=kT/(Avogadro's number)

14. Energetic contribution of a hydrogen bond: ~6-20 kJ/mole (BNID 103914, 103913, 101067, 101069, 101068, 101070)
   9 kJ/mole Strength of hydrogen bond in water (BNID 103914)

Measurement Method: An extensive series of over 400 molecular dynamics simulations with an aggregate length of over 900 ns was analyzed using an analysis technique in which hydrogen bond (HB) breaking is interpreted as an Eyring process, for which the Gibbs energy of activation ΔG can be determined from the HB lifetime.

15. **DG** of ATP hydrolysis under physiological conditions in E. coli ~50 kJ/mole (BNID 101701)
   PMID 949233

Measurement Method: calculated from the equation: delta G (Phos) = delta G (at standard conditions) + RT ln([ATP]/[ADP][Pi]), where R is the gas constant, T = temperature, and -30.5 kJ/mol was used for delta G at standard conditions.
Comments: The number has been converted from positive to negative so that it describes the Gibbs free energy of hydrolysis as opposed to the phosphorylation potential. The E. coli was growing in the presence of oxygen. See reference for more details.

16. Number of ATP molecules required to make an E. coli cell on minimal medium and glucose under Aerobic conditions \(-10^{10}\) ATP (BNID 101981)


Measurement Method: YATP(max) was determined by measuring the rate of oxygen consumption(QO2) and growth rate(D), and calculating the value of mol ATP formed per mol oxygen (N) and the energetic maintenance cost (M). The equation QO2 = D/YATP(max)*N + M/N was used.

Comments: This number is calculated from the value for YATP(max) of 13.9 in the reference. YATP(max) is the number of grams of cells (dry weight) produced per mol of ATP corrected for cellular maintenance energy costs. 1/13.9 gives 0.0719 mol of ATP per gram of cells. One E.coli cell weighs 0.28pg (dry wt), so there are 3.57 trillion cells/gram. 0.0719*6.02E23/(3.57E12 cells/g (dry wt)) gives 12.1 billion ATP/cell.

Number of ATP molecules to make an E. coli cell on minimal media and glucose under anaerobic conditions \(1.64\times10^{10}\) ATP (BNID 101983)


Comments: This number is calculated from the value for YATP(max) of 10.3 in the reference. YATP(max) is the number of grams of cells (dry weight) produced per mol of ATP corrected for cellular maintenance energy costs. 1/10.3 gives 0.0971 mol of ATP per gram of cells. One E.coli cell weighs 0.28pg (dry wt), so there are 3.57 trillion cells/gram. 0.0971*6.02E23/(3.57E12 cells/g (dry wt)) gives 16.4 billion ATP/cell.

ATP requirement for growth on acetate and minimal media under aerobic conditions \(2.48\times10^{10}\) ATP (BNID 101982)


Measurement Method: YATP(max) was determined by measuring the rate of oxygen consumption(QO2) and growth rate(D), and calculating the value of mol ATP formed per mol oxygen (N) and the energetic maintenance cost (M). The equation QO2 = D/YATP(max)*N + M/N was used.

Comments: This number is calculated from the value for YATP(max) of 7.1 in the reference. YATP(max) is the number of grams of cells (dry weight) produced per mole of ATP corrected for cellular maintenance energy costs. 1/7.1 gives 0.1408 mol of ATP per gram of cells. One E.coli cell weighs 0.28pg (dry wt), so there are 3.57 trillion cells/gram. 0.1408*6.02E23/(3.57E12 cells/g (dry wt)) gives 24.8 billion ATP/cell.

ATP requirement for growth and Yatp under different nutritional conditions Table Link - http://tinyurl.com/qjojft (BNID 101637)

Ref: Byung Hong Kim, Geoffrey Michael Gadd. Bacterial Physiology and Metabolism. Cambridge University Press.


Comments: After consulting primary sources, it appears that for columns E and F, and possibly G, the energy requirement for mRNA turnover was already included in the RNA cost, and so has been counted twice in this table. This would reduce the "Total" by 1.39 mmol of ATP for these columns, which would cause a very slight change in Yatp.

Diffusion and catalysis rate

17. Diffusion coefficient of dextran (40kDa) in Drosophila cytoplasm 17.6+-1.8 \(\mu\)m\(^2\)/sec

(BNID 100198)

Measurement Method: Fluorescent dextran injected into Drosophila embryo, confocal imaging, and fitting with 3D diffusion model

7.7±2.5 μm²/sec GFP Diffusion coefficient in E. coli cytoplasm (BNID 100193)

Measurement Method: FRAP and photoactivation

Time for protein to diffuse across E. coli cell ~0.1 sec (BNID 104667)
Ref: Calculated manually. Please see measurement method.

Measurement Method: According to equation: t diffusion≈X²/2D. Taking X, distance to be traveled, as 1um and D, diffusion coefficient, as 100um²/sec. D calculated from D=KBT/6/π/η/R where R=2.5nm, typical protein diameter. KB=Boltzmann's constant, η(viscosity of the medium, taken as 0.001 Pa*sec for water at ~300 Kelvin, T=temperature in degrees Kelvin, π≈3.14. (1.380 6504(24)×10^-23 Kgm²/sec²·2K^-1·300K)/(6·3.14·0.001Kgm^-1·sec^-1·2·5·10^-9 m)=8.8×10^-11m²/sec=88um²/sec=~100um²/sec

Comments: Diffusion coefficient of 100 um²/sec refers to water. In cytoplasm D is smaller than 100um²/sec as there are solutes, in the range of 5-15 um²/sec (BNIDs 100193, 100198).

Taking D as 10 um²/sec, actual time of diffusion would be (1um)²/(2·10)um²/sec=~0.1 sec

~5 sec for hemoglobin to traverse erythrocyte (BNID 104677)
Ref: Calculated according to erythrocyte diameter. Please see Measurement Method

Measurement Method: Calculated with equation d²=2Dt where d=distance to pass, length of erythrocyte, 10um. D of hemoglobin in erythrocyte cytoplasm is lower than in water due to presence of solutes, in the range of 5-15 um²/sec (BNIDs 100193, 100198). Taking D as 10 um²/sec, actual time of diffusion would be (10um)²/(2·10)um²/sec=~5 sec BNID 104106 for diffusion time in water medium

D (metabolite) ~500 mm²/sec
Diffusion coefficient of sucrose in water 520 μm²/sec (BNID 100614)
Ref: S. Vogel Life's Devices (1988)

Diffusion coefficient of glucose in water 600 μm²/sec (BNID 104089)

Comments: Table gives Molecular/Ionic weight and Diffusion coefficients of various substances in specified temperatures

18. Diffusion limited on-rate for a characteristic protein ~10^8-10^9 1/sec/Molar (BNID 103916)

Comments: There is an upper limit to Kcat/Km, imposed by the rate at which E and S can diffuse together in an aqueous solution. This diffusion controlled limit is 10⁸ to 10⁹ M⁻¹·s⁻¹·1, and many enzymes have a kcat/Km near this range

Division, Replication, transcription, translation rates

19. Exponential cell cycle time:
E. coli ~30 minutes (BNID 103890)
**Comments**: Table gives Protein/mass, RNA/mass, DNA/mass, cell no./mass, protein/genome, RNA/genome, origins/genome, protein/origin and more parameters at 5 different doubling times

Yeast: ~100 minutes (BNID 100270)


**Comments**: Can reach ~70 minutes under ideal conditions (see BNID 101747)

Population doubling time, percent budded cells, and mean cell volume for different batch culture media 71-477 min (BNID 101747)


**Measurement Method**: The mean size and percentage of budded cells of a wild-type haploid strain of *Saccharomyces cerevisiae* grown in batch culture over a wide range of doubling times (tau) have been measured using microscopic measurements and a particle size analyzer.

**Comments**: Shortest generation time 71 min on YEP+fructose medium Longest generation time 477 min on EMM+acetate+phthalate

Human cell line (Hela): 16.2 h (BNID 103804)


**Measurement Method**: Hypergravity cell culture, [3 H] thymidine incorporation, Cell cycle analysis, RNA blot hybridization (Northern analysis)

**Comments**: Research checked difference between HeLa cells in hypergravity conditions and control.

Doubling time of cancerous cell lines 17-80 hours (BNID 100685)

**Ref**: [http://dtp.nci.nih.gov/docs/misc/common_files/cell_list.html](http://dtp.nci.nih.gov/docs/misc/common_files/cell_list.html)

**Measurement Method**: In vitro

**Comments**: Information on generation time, in hours, of the NCI set of 60 cancerous cell lines. Range 17-80 hours

Doubling time of human cancerous cell lines 13-80 hours (BNID 103994) Table link - [http://tinyurl.com/clejlo](http://tinyurl.com/clejlo)

**Ref**: Patent - JOINT USE OF SULFONAMIDE BASED COMPOUND WITH ANGIOGENESIS INHIBITOR-OWA, OZAWA, and SEMBA 20.06.2007 link - [http://tinyurl.com/cd9eko](http://tinyurl.com/cd9eko) page 38 table 1

**Measurement Method**: Cells of these cell lines were seeded in 96-well microplates (flat-bottomed) (50 Pl/well) at the cell numbers indicated in Table 1. After 24 hours, a 3-fold dilution series of each compound was added (50 Pl/well). After 72 hours, WST-8 (10 Pl/well) was further added and the absorbance at 450 nm was measured. Additional information under table.

**Comments**: Information on generation time, in hours, of 36 cancerous cell lines. Range 13-80 hours.

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20. **Average replication rate of T7 bacteriophage DNA in *E. coli* 220+-80 nucleotides/sec** (BNID 103995)


**Measurement Method**: in vitro, single molecule measurements. The 5’ end of one strand of a 48.5 kilobase (kb)-long duplex lambda phage DNA molecule is attached to the bottom surface of a glass flow cell using a biotin–streptavidin linker. The opposite 3’ end bears a digoxigenin moiety and is linked to a bead that is 2.8 microm in diameter and coated with anti-digoxigenin antibody

**Comments**: These values are in agreement with those observed in a previously published single-molecule experiment: Wuite, G. J. L., Smith, S. B., Young, M., Keller, D. & Bustamante, C. Single-molecule studies of the effect of template tension on T7 DNA

DNA synthesis rate of E. coli DNA polymerase III holoenzyme ~750nts/sec (BNID 104120)


Measurement Method: In vitro. The reconstitution approach was taken to study which subunits of the heterotrimer core polymerase (alpha, epsilon, theta) participate in the highly processive replication of long DNA templates by DNA polymerase III holoenzyme (holoenzyme).

Comments: This value is consistent with the rate of fork movement in E. coli. This rapid rate results from the high processivity of holoenzyme, which extends a chain for several thousand nucleotides without dissociating from the template even once.

DNA replication rate in human 33 nts/sec (BNID 104136)


Measurement Method: Replication fork speed of ~ 2000 base pairs/min=2000/60 base pairs/sec=33 base pairs/sec

Comments: Article also gives info on distance between origins of replication: For a human cell, the approximately 3,000,000 kb genome is replicated in about 8 h. Since the rate at which replication forks plow through chromatin is no greater than ~ 2 kb/min, bidirectional orgins would need to be spaced at roughly 2000 kb intervals to finish replication in the allotted time.

21. Rate of transcription by RNA polymerase ~70 bp/sec

70 nt/sec for E. coli (BNID 100060)


~71.6 nt/sec mammalian tissue culture cell (BNID 100662)


Measurement Method: parameter optimization of FRAP and photoactivation data in U2OS 200 copy gene array. LacI gene integrated into Human U2OS osteosarcoma cells

Comments: See also average polymerase velocity and pause residence time. YFP-Pol II is amanitin resistant allele.

22. Rate of translation by the ribosome ~20 aa/sec

12-21 aa/sec for E. coli (BNID 100039)


Comments: lower value is for slow division rate (100 minutes) and higher value is for fast division rate (24 minutes)

Genomes, Mutation and error rates

23. Genome size:

E. coli ~4×10^6 bp (BNID 100269)
Comments: The 4,639,221-base pair sequence of Escherichia coli K-12 is presented.

S. cerevisiae ~12×10^6 bp (BNID 100459)
Ref: http://www.yeastgenome.org/cache/genomeSnapshot.html
Comments: The Budding yeast genome size is 12,156,676 base pairs as of May 6th 2009

C. elegans ~100×10^6 bp (BNID 101363)
Comments: The complete Caenorhabditis elegans genome sequence, fully contiguous telomere to telomere totals 100,291,840 bp.

D. melanogaster ~120×10^6 bp (BNID 100199)

Arabidopsis ~157×10^6 bp (BNID 104000)
Ref: Bennett MD, Leitch IJ, Price HJ, Johnston JS. Comparisons with Caenorhabditis (approximately 100 Mb) and Drosophila (approximately 175 Mb) using flow cytometry show genome size in Arabidopsis to be approximately 157 Mb and thus approximately 25% larger than the Arabidopsis genome initiative estimate of approximately 125 Mb Ann Bot (Lond). 2003 Apr 91(5):547-57. PMID 12646499

Mouse ~3×10^9 bp (BNID 100305)
Ref: http://wormlab.caltech.edu/briggsae/genomeSize.html

Human ~3×10^9 bp (BNID 100396)
Comments: Value refers to Euchromatic genome. For heterochromatic genome see BNID 101484

24. Mutation rate in DNA replication ~10^{-9} per bp in E. coli
5.4E-10 mutation/bp/replication (BNID 100263)
Ref: Drake JW. A constant rate of spontaneous mutation in DNA-based microbes. Proc Natl Acad Sci U S A. 1991 Aug 15 88(16):7160-4 Table 1 PMID 1831267
Measurement Method: Value is mean of two strains-hisGDCBHAFE (one measurement) and LacI (2 measurements)

Mutation rate in DNA replication 5×10^{-11} per base pair per replication in human (BNID 100414)

25. Error rate in transcription ~10^{-4} per nucleotide (BNID 103453)
Comments: This level of accuracy is lower than that of DNA replication and similar to that of translation.

Error rate in translation ~10^{-3}-10^{-4} per amino-acid (BNID 103454)
Primary Source: (1) Bouadloun, D. Donner and C.G. Kurland, Codon-specific missense errors in vivo, EMBO J. 2 (1983), pp. 1351–1356. PMID 10872330 AND (2) P. Edelmann and J. Gallant, Mistranslation in E. coli, Cell 10...
(1977), pp. 131–137. PMID 138485

**Comments:** This level of accuracy is lower than that of DNA replication and similar to that of transcription