



Cell extract gels as an example of active matter

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Abstract

Cell lysates (cellular extracts) constitute a perfect imitation of the intracellular environment that can provide insight into cellular response to external stimuli. However, most of the presented results are performed for diluted lysates that do not reflect the actual properties of a crowded cellular environment. Here, we report for the first time the measurement of the viscosity and shear storage modulus of highly concentrated *Escherichia coli* (*E. coli*) lysates with and without adenosine triphosphate (ATP). By cleavage of DNA content, we showed the value of shear storage modulus G' decreases by 19–31% in comparison to control samples. The addition of molecules that provides energy (ATP) allowed to rebuild the structure of the lysate by reversibly increasing viscous properties over elastic ones. When the energy delivered in the form of ATP is consumed by the unliving bacterial lysate, the system returns to its initial state.

Keywords *Escherichia coli* · *E. coli* · Lysate · Viscoelasticity · Adenosine triphosphate · ATP · Shear modulus

Introduction

A concept of active matter—the ubiquitous nonequilibrium condensed system—has been developed for many years in the context of both living and nonliving matter (Marchetti et al. 2013; Palacci et al. 2010; Paxton et al. 2004; Parrish and Hamner 1997; Serra-Picamal et al. 2012). The active matter systems are composed of self-driven units, active particles, each capable of converting stored or ambient free energy into systematic movement (Schweitzer 2007). This energy-related concept is utilized in all biological systems on different scales, starting from cells (Kemkemer et al. 2000) or bacterial suspensions (Dombrowski et al. 2004) and with shoals of fish or flocks of birds (Ballerini et al.

2008). In this paper, we present results on highly concentrated *Escherichia coli* (*E. coli*) lysate as an alternative system, in which the consumption and dissipation of energy in the form of added adenosine triphosphate (ATP) allow the system to change its viscous and elastic properties.

Foffano et al. (2012) simulated macroscopic shear experiments in active nematics and compared them with microrheology simulations where a spherical probe particle was dragged through an active fluid. They showed that effective viscosity, rather than being solely a property of the active fluid, was affected by the way chosen to measure it and strongly depended on details such as the anchoring conditions at the probe surface and on both the system size and the size of the probe particle, whereas the rheological studies performed by Liu et al. (2019) using *E. coli* (as model active swimmers), in a microfluidic viscometer under different degrees of confinement, revealed a strong confinement effect on dilute bacterial suspensions: the apparent viscosity of suspensions was reduced by a factor of three when the confinement scale decreases from 60 μm down to 25 μm . They demonstrated that the effect of confinement is directly linked to the motility of bacteria. The studies of Martinez et al. (2020), which used bulk rheometry and particle-tracking rheoimaging, showed how suspending self-propelled “pushers” like *E. coli* in a liquid influences system size in bacterial suspensions. Above the critical bacterial volume fraction needed to decrease the viscosity to zero, $\phi_c \approx 0.75\%$, large-scale

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collective motion emerged in the quiescent state, and the flow became nonlinear. They confirmed a theoretical prediction that such instability should be suppressed by confinement. In turn, Lin et al. (2007) measured the linear and nonlinear viscoelastic properties of isotropic solutions of purified microtubules, as well as networks permanently cross-linked with biotin NeutrAvidin. They showed that in the linear regime both solutions and networks are soft elastic materials with elastic moduli on the order of a few pascals. The elastic moduli showed a power law dependence on tubulin concentration, c_T , with $G' \sim c_T^\nu$, where $\nu \approx 1.4$ for solutions and increased slightly to $\nu \approx 1.6 - 1.8$ for networks. At large deformations, they observed concentration-dependent yield stress. Next, Arevalo et al. (2010) investigated the size-dependent rheological response of branched type I collagen gels and observed that when subjected to a shear strain, the highly interconnected mesh dynamically reorients, resulting in overall stiffening of the network. When a continuous shear strain to a collagen network was applied, the local apparent modulus, in the strain-stiffening regime, strongly depended on the gel thickness. They also demonstrated that the overall network failure was determined by the ratio of the gel thickness to the mesh size. Pelletier et al. (2009) demonstrated that the viscoelastic properties of microtubules, as reported from two-point microrheology, were in line with the macroscopic measurement at high frequencies, but appeared to show a discrepancy at low frequencies, at time scales on the order of a second. A composite of filamentous actin (F-actin) and microtubules had viscoelastic behavior between that of F-actin and pure microtubules. They further showed that the Poisson ratio of the composite, measured by the length scale-dependent two-point microrheology, was robustly smaller than that of the F-actin network at time scales $\tau > 1s$, suggesting that local compressibility was conferred by the addition of microtubules to the F-actin network. Sato et al. (1988) measured the elasticity and viscosity of brain tubulin solutions under various conditions with a cone and plate rheometer using both oscillatory and steady shearing modes. Microtubules composed of purified tubulin, purified tubulin with taxol, and 3 times cycled microtubule protein behaved as mechanically indistinguishable viscoelastic materials. Microtubules composed of pure tubulin and heat-stable microtubule-associated proteins were also similar but did not recover their mechanical properties after shearing like other samples, even after 60 min. All of the other microtubule samples were more rigid after flow orientation, suggesting that the mechanical properties of anisotropic arrays of microtubules could be substantially greater than those of randomly arranged microtubules.

In vivo experiments (Weber et al. 2010), as well as measurements on disrupted cells (Kong et al. 2009), suggest that the cytoplasm of the living cell is a non-Newtonian,

viscoelastic medium whose properties are determined by the DNA. Moreover, a recent report (Parry et al. 2014) also suggests that the transport properties (viscosity or fluidity) of the intracellular environment can also be regulated by adenosine triphosphate (ATP)-dependent processes. Changes of rheological properties such as viscoelasticity are connected to the changes of structure of the system; in consequence, it is connected to the movement of molecules in the system forced by the addition of energy (ATP).

Parry et al. (2014) have shown that the fluidity of the cytoplasm of *E. coli* drops down in ATP-depleted cells. The effect is manifested as a decrease in the mobility of protein aggregates. In the case of untreated cells, the aggregates (approximately 60 nm in size) diffuse freely along with the whole cell. In ATP-depleted cells, the mobility of the aggregates is significantly reduced. The mobility decreases with the size of aggregates and the largest probes are found to be almost immobile. The experiment of Parry et al. (2014) suggests that monitoring the motion of the proteins inside the living cell can be used to probe the structural changes of the cell interior in the presence of external stimuli. However, the information about the mechanical response is limited to the length scale that corresponds to the size of the protein. In fact, length scale is spatially constrained to the region in which the protein's motion occurs. To explore the average response of the intracellular matrix, it would be more convenient to have an experimental model system that can be investigated in the macroscopic length scale, for example, by means of rheometry.

Lysate has all components of the cytoplasm of living cells and is obtained at a macroscopic volume of several milliliters, which allows to perform rheological measurements. Therefore, the cell lysate is not only an example of active matter but also constitutes a perfect in vitro model to study the average viscoelastic response of the cytoplasm.

Usually, the lysate is obtained using a procedure in which the bacteria are suspended in a buffer or in a culture media. To disrupt the cells, the bacterial culture is treated with a surfactant (i.e., sodium dodecyl sulfate (SDS)), enzymes (i.e., lysozyme), or ultrasonic waves. These procedures require the dilution of cells, resulting in the dilution of the obtained matrix. Consequently, such lysates do not resemble the intracellular environment. Therefore, the dilution of the cell culture and the lysate should be minimized, to obtain conditions that are the same as those in living cells.

Up to date, the rheological measurements for bacterial lysates have been conducted in several reports (Stephenson et al. 1991; Wright et al. 2001; Ciccolini et al. 1999; Malik et al. 2006; Levy et al. 1999; Ciccolini et al. 1998). These measurements have been used to monitor the kinetics of the lysis process (Ciccolini et al. 1999) or the DNA content after antibiotic treatment (Malik et al. 2006). To the

best of our knowledge, there are no examples of studies of the rheological properties of highly concentrated cell extracts—the extracts that do not contain any additional water apart from that enclosed within the cells, followed by the examination of the active response to external stimulants. In this paper, we report our investigation of the rheological properties of a highly concentrated cell lysate and the influence of DNA content on rheological properties of the lysate. We also demonstrate how the concentration of the lysate modifies the viscoelastic properties of the suspension. Finally, we show that the addition of ATP temporarily reduces the shear storage modulus of the lysate. Thus, this result suggests that the biological functions of the constituents are maintained during the lysis procedure.

Methods

Culture and growth conditions

A turbid overnight culture of *E. coli* K-12 (ATCC 10798) was used to inoculate fresh lysogeny broth (LB Miller, A&A Biotechnology) at a proportion of 200 μ l culture to 20 ml LB. The freshly inoculated broth was incubated at 37 °C in Erlenmeyer flasks in a shaker incubator (infors-ht minitron, 180 rpm) until the culture reached OD600 of around 1 (after about 3–4 h; estimated on the basis of the growth curves—Figure S1 Cells were collected by centrifugation, Sorvall RC 6 Plus, rotor SLC 3000, 5000 rpm (4400 g)), at 20 min at 4 °C. The supernatant was removed from the sample, and the collected pellet was suspended in a phosphate buffer (pH = 7.2) and again centrifuged. The collected pellet was stored at –20 °C before further processing. Usually, 10 l of bacterial culture resulted in around 20 g of the bacterial pellet.

Lysis

Lysis was carried out using cell disruptor TS2/40 (Constant Systems Ltd., UK). If not stated otherwise, the lysis procedure was as follows. The obtained pellet stored at –20 °C was defrosted and then, without further dilution, lysed five times. We used the cell disruptor operating under a pressure of ~206 MPa (30 kpsi) with the ONE SHOT head adaptor. The adaptor circumvented the need for sample dilution. The 5-fold lysis repetition was sufficient to obtain nearly 100% lysis efficiency. The obtained lysate was stored frozen at –20 °C in 50-ml falcon vials.

Sample preparation

Before the first use, the as-prepared lysate was once again centrifuged (5000 rpm, 5 min, 20 °C) in order to

remove the excess of the LB left after the process of lysis, the air bubbles, and the foam formed during lysis. Lysate dilutions were prepared using a Gilson Microman pipette for highly viscous liquids. This allowed us to express the concentration in terms of volume fraction. For the rheological measurements, we used 1 ml of the sample containing either pure lysate or lysate diluted to the desired volume fraction. For the diluted samples, we used a phosphate buffer (pH = 7.2) as a solvent. The buffer prevented the lysate constituents from, e.g., incorrect folding in the case of proteins.

DNA cleavage: DNA cleavage was performed after mechanical lysis. Pure lysate (850 μ l, $\phi = 1$) was mixed with 50 μ l of DNAses I (10000U, A&A Biotechnology, Poland) and 100 μ l of reaction buffer (A&A Biotechnology, Poland). The mixture was incubated for at least 1 h and then analyzed with a rheometer. The control samples were prepared in the same manner, but without DNAses. The final volume fraction of the samples was equal to $\phi = 0.85$.

Addition of ATP: Samples with adenosine triphosphate (ATP) were prepared after mechanical lysis. Nine hundred fifty microliters of the pure lysate was mixed with 50 μ l of the solution of ATP (50 mg/ml) dissolved in the phosphate buffer (pH = 7.2).

Rheology

For rheological measurements, we used the rotational rheometer Kinexus Pro (Malvern Instruments Ltd., UK). The measurements were performed at 25 °C. A steel cone-plate geometry, with a diameter of 50 mm, an angle of 1°, and a 30- μ m gap, was used to perform rotational measurements giving the shear viscosity of the extract. A cone-plate geometry or a plate-plate geometry is the most common geometries used in the rheological characterization of viscoelastic biological samples such as lysate (Newton et al. 2017). The experiments were done in shear stress controlled mode and free shear rate. We calculated the “zero shear” viscosity by the extrapolation of the measured shear viscosity to the zero shear rate.

We performed oscillatory measurements using a steel plate-plate geometry, a diameter of 20 mm, and a 100- μ m gap. The utilization of this geometry allowed us to decrease the amount of lysate necessary to one measurement (both cone-plate and plate-plate geometries are used in rheological measurements (Kinloch et al. 2002; Ewoldt et al. 2008)). For all samples, except those for the ATP time series, the oscillatory measurements consisted of two types of measurements. The first one was an amplitude sweep test to find the linear viscoelastic region (LVER). The second was a frequency sweep test to describe the rheological type of the samples. In the amplitude sweep test, we measured the shear storage and the shear loss moduli

(G' and G'') dependence on the strain (range from 0.01 to 100% at a frequency of 1 Hz). This test showed us how big deformation we could apply to the sample without destroying its structure. Using the amplitude of strain from LVER, found in the amplitude sweep test, we performed the frequency sweep test (range 10 – 0.1s⁻¹). From the frequency sweep test we obtained $G^* = G' + iG''$ and δ (phase angle) at different time scales. These measurements allowed us to classify the lysate samples as a gel because δ was found to be independent of the frequency in the whole range of the oscillations.

Results and discussion

The interior of a bacterial cell is a highly crowded environment—up to 30% of its volume is occupied by macromolecules. In order to mimic such a crowded environment, composed of all the biologically relevant constituents, we performed one-shot lysis of the cells. For the lysis, instead of cell suspension, we used bacterial paste—a non-diluted bacterial pellet. This procedure allowed us to obtain a lysate with a very small additional amount of water no more than 30%. The presence of additional water could affect the real concentration of macromolecules inside the lysate and alter the absolute values of viscosity and the storage moduli. This value can differ between lysate batches. To minimize the variance between samples whenever it was possible, we expressed our experimental results as relative values with respect to the values obtain within one batch of lysate. Alternatively, we compared the results to the control samples prepared from the same batch. The concentration of the lysate was expressed as the volume fraction of the lysate (not macromolecules), ϕ .

We performed measurements of the shear viscosity for a pure, non-diluted lysate ($\phi = 1$) and for dilutions with buffer. The samples containing pure lysate were found to be non-Newtonian over the whole range of applied shear rates. A non-Newtonian response was observed also for the diluted samples (as indicated in Fig. 1) and disappeared for volume fraction below $\phi = 0.1$. All viscoelastic samples exhibited a gel-like behavior when the measured phase angle was independent of the frequency (see Fig. 2 and Figures S2–S5).

For solutions at volume fraction $\phi < 0.1$, the viscous response was measured. The measurements indicated that the viscosity η was comparable to that of water η_0 reaching $\eta \approx 2\eta_0$ at $\phi = 0.1$. The plots of viscosity against shear rate showed that above $\phi = 0.1$ the samples could not be analyzed as a Newtonian fluid. Oscillatory measurements of the shear storage modulus G' performed for samples at volume fraction $\phi < 0.3$ were unsuccessful because the

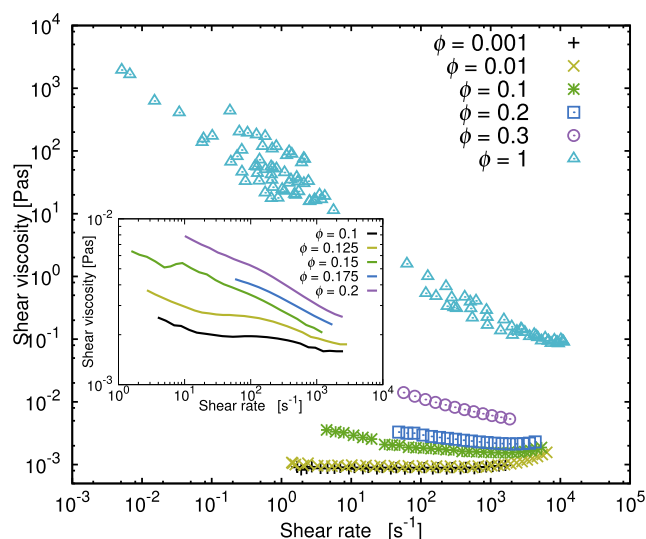


Fig. 1 The plot showing shear viscosity of the *E. coli* lysates at various volume fractions ϕ . The volume fraction is not related to the fractions of macromolecules but to the overall amount of cell extract used. $\phi = 1$ means that as-prepared extract was used without further dilution. Samples below $\phi = 0.1$ exhibited Newtonian behavior, where the shear viscosity was not dependent on the shear rate. Samples with a higher volume fraction ($\phi \geq 0.1$) exhibited non-Newtonian response on the rate of shear. Inset shows shear viscosity measured for the range of concentrations from 0.1 to 0.2

inertia of the geometry used in the experiments was greater than the elastic response of the sample. For volume fraction $\phi > 0.3$, the samples were clearly viscoelastic and the shear storage modulus increased with the extract volume fraction.

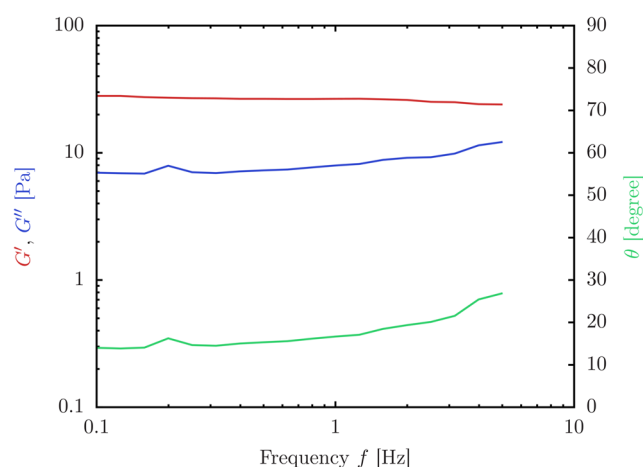


Fig. 2 The plot shows the dependences of G' , G'' , and δ on the frequency of oscillations for volume fraction $\phi = 0.70$. Data shown in both plots were acquired for the mechanically obtained lysate, according to the procedure described in the main text. The measurement was performed at 25 °C. As indicated in the figure, there is no clear association between the phase angle and frequency of oscillations. These results suggest that the samples pose a gel-like structure

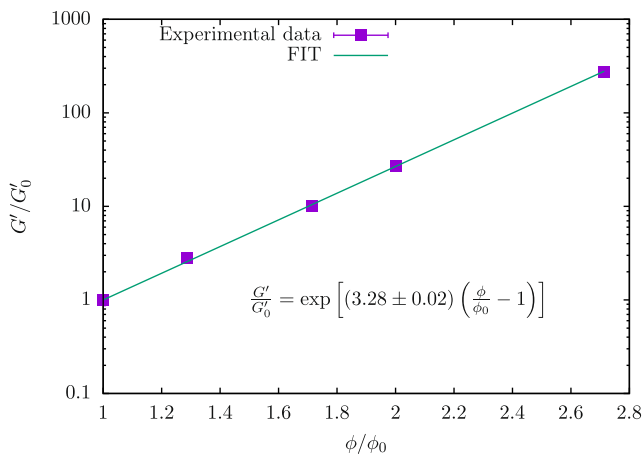


Fig. 3 The shear storage modulus, $\frac{G'}{G'_0}$, as a function of volume fraction $\frac{\phi}{\phi_0}$ of the lysate. The figure shows the shear storage modulus obtained from macroscopic measurements. The modulus was calculated from the linear viscoelastic region. It was estimated from the amplitude sweep test; a range of shear strain from 0.01–1%, frequency $f = 1$ Hz. The lysate exhibits non-Newtonian character (at volume fraction $\phi > 0.3$). The solid line correspond to the fit of Eq. 1 to the macroscopic data with parameters: $B_\omega = 3.28 \pm 0.02$

According to Eq. (1), we express G' as an exponential function of ϕ , where ϕ_0 is the lowest volume fraction and G'_0 is the lowest shear storage modulus at which viscoelastic response was observed. Data are depicted in Fig. 3

$$\frac{G'}{G'_0} = \exp \left[B_\omega \left(\frac{\phi}{\phi_0} - 1 \right) \right] \quad (1)$$

The fluctuation of G' in different batch and this same batch are plotted in Fig. 4. The presented values of storage modulus in the same batch are repeatable. Since each batch

Fig. 4 Comparison of the shear storage modulus G' 3 different series and in every series for 3 samples of pure lysate $\phi = 1$

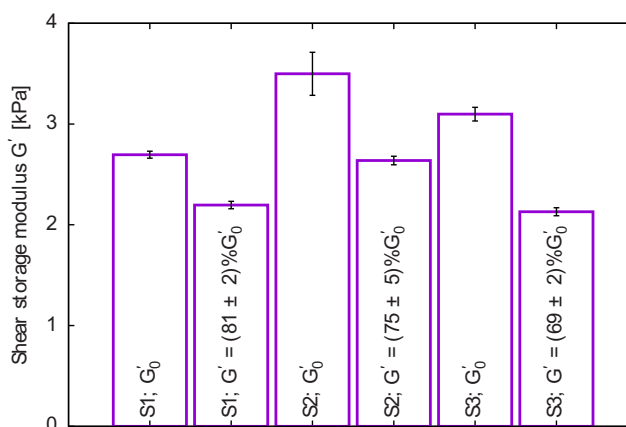
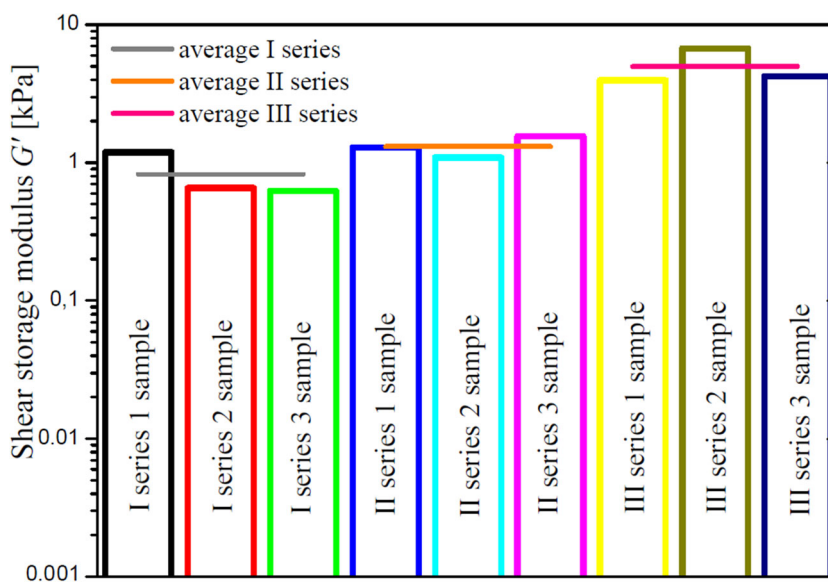


Fig. 5 Comparison of the shear storage modulus G' for DNase treated lysates and control samples. A decrease ranging from 19 to 31% was observed for samples S1 to S3 treated with DNases (used for cutting DNA chains inside the lysate)

was prepared from a different bacterial culture, storage modulus in different batches differs significantly. For this reason, we compared the normalized values so that other pairs of lysates could also be compared.

Although we knew the viscoelastic properties of the tested system and we were able to determine them not only qualitatively but also quantitatively, the key problem was to determine what can cause such a rheological property of the system. After analysis of the composition of the sample, it was assumed that the elasticity of the lysate gel originates from its constituents such as DNA. To verify to what extent DNA influences the viscoelastic properties of the cell extract, we performed a DNA cleavage process that was followed by tests to estimate the influence of DNA on the shear storage modulus of the cell extracts. The

experiment involved the biochemical removal of the given components from the extract.

After the mechanical lysis, we mixed the extract with a highly concentrated solution of type I DNases that randomly cut the DNA into smaller fragments. This treatment should cause a decrease in the overall storage modulus of the sample. After incubation with DNases I, the treated lysate was compared with untreated extract at the same volume fraction. DNA cleavage led to a decrease (from 19 to 31%) in the shear storage modulus of the extract (see Fig. 5). All samples exhibited a gel-like behavior (see Figs. 6, 7 and S6–S9). These results indicate that the presence of entangled DNA makes the cellular extract viscoelastic. According to the paired *t* test results, differences in the shear storage modulus of DNases treated and the untreated lysate are considered to be statistically significant. Due to the crowding of the system, we cannot determine the efficiency at which the enzyme cleaved DNA. Therefore,

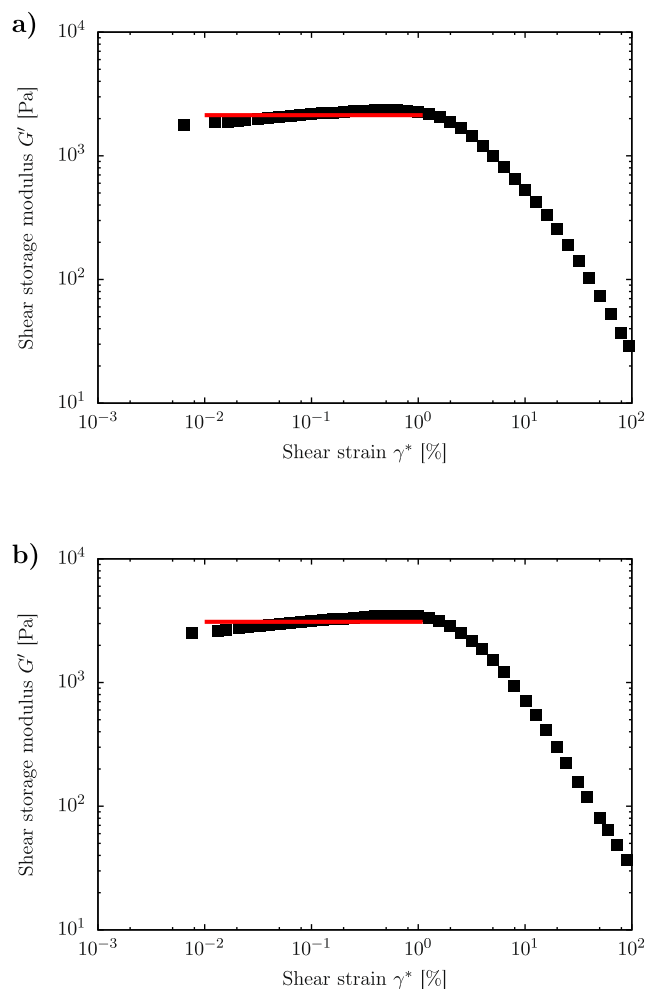


Fig. 6 The plot shows the dependencies for sample 3 with DNases and control sample. Panel **a** shows dependencies of G' on strain of oscillation in sample 3. The red line shows the LVER. Panel **b** shows the result for control sample

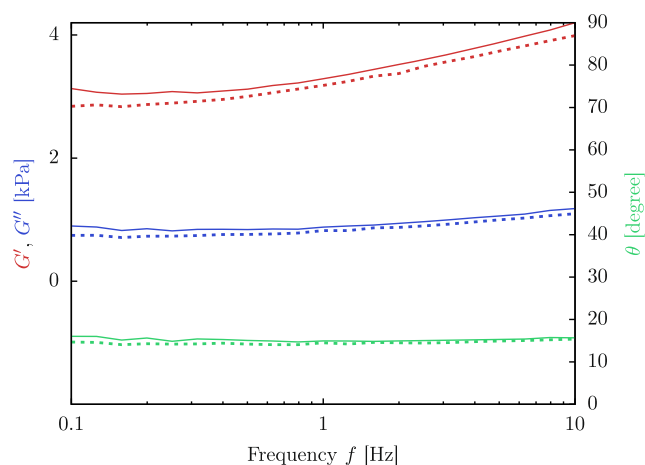


Fig. 7 The plot shows results for sample 3 (solid line) and control sample (dotted line) of G' , G'' , and δ on the frequency of oscillations

the observed drop confirms only the qualitative and non-quantitative effect of DNA on the viscoelastic properties of the sample.

Although the cellular extracts are composed of the same constituents (DNA, proteins, sugars, and amino acids), they differ from the cytoplasm of living cells in one important aspect. The lysates are nothing more than disrupted cells, which bio-synthetic machinery is stopped at the moment of cell disruption—this also includes the ATP production machinery. Therefore, in the next step, we wanted to answer the question whether the structure of a nonliving bacterial lysate may be converted due to the addition of molecules that provides energy in all biological processes. We expected that the lack of ATP should lead to an increase of shear storage modulus of the lysate, analogously to what was observed for living cells by Parry et al. (2014). On the other hand, the addition of ATP to the cell extract should result in a decrease in the shear storage modulus. This might be explained by the fact that in the presence of the source of chemical energy (ATP), the lysate constituents can still perform their functional work.

We performed measurements of the shear storage modulus as a function of time after the injection of ATP to cell extracts (see “Sample preparation”). Each time point represents a single sample of lysate to which a solution of ATP had been added and stirred. We performed three experimental series (series I, II with ATP and III series with buffer/ADP). For each series, the same experimental procedure was maintained. Within an experimental series, the same volume of ATP/ADP/buffer was added to all samples at the same time point. For every sample, we added the same volume of lysate and buffer, to be sure that we compare samples with the constant volume fraction of the lysate. After incubation at room temperature in sterile conditions, the sample was placed on the rheometer and

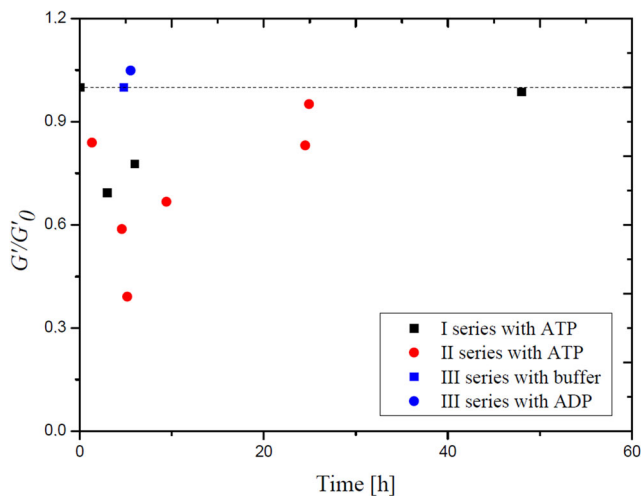


Fig. 8 Shear storage modulus G'/G'_0 as a function of time after the addition of ATP, ADP, or buffer to the lysate. Each point corresponds to a single sample for which the oscillatory measurements were performed. All samples in each series were prepared from the same batch of the lysate (each series from a different batch), and the solution of ATP, ADP, or pure buffer was added to all of them at the same time and stirred. Then, the samples were measured at the intervals shown in the plot. After about 6 h, a drop of the storage modulus was observed for samples with the addition of ATP. The dashed line represents the initial value of the storage modulus measured for the first sample

oscillatory measurements were performed. We observed a significant decrease of the shear storage modulus G' (almost 3 times) during the first 6 h after ATP injection. After 6 h, the G' started to increase and finally (after 2 days) reached its initial value. This suggested that the system had consumed all the introduced ATP and returned to its initial state. Figure 8 depicts the kinetics of ATP consumption on changes of the storage modulus.

The complex character of the lysate causes that it is impossible to pinpoint a single constituent that interact with ATP leading to decrease of G' . Therefore, estimation of time scales related to ATP consumption become difficult. Instead, it is instructive to discuss the rates of ATP consumption. In living bacterial cells, the rate of ATP consumption is about 100 mmol of ATP per hour per gram of dry weight (Milo and Phillips 2015). For our samples, that value translates into ~ 3.5 mmol/h. Assuming that all of 5 μmol of ATP available in the samples was consumed within 48 h, the average rate of ATP consumption was approximately 0.1 $\mu\text{mol/h}$. This value is about 4 orders of magnitude lower than expected for living cells.

Conclusions

In this study, we investigated the rheological properties of a highly concentrated *E. coli* extract. First, we analyzed the dependence of the shear viscosity on the volume fraction of

the lysate. We found that at a wide range of volume fraction of the cellular extract behaved as a gel-like non-Newtonian fluid. The extract only behaved as a Newtonian liquid for extract volume fraction $\phi < 0.1$. The elastic response (shear storage modulus) of the lysate was observed only at volume fraction $\phi \geq 0.3$. We described the shear storage modulus by Eq. 1. The shear storage modulus of the extract depends on DNA chains. The samples with enzymatically cleaved DNA exhibit a decrease in the value of the shear storage modulus by 19 to 31% of the original value. Finally, we monitored the kinetics of ATP consumption in the lysate. A few hours after the addition of ATP, the shear storage modulus of the lysate dropped down almost 3 times. After 2 days, the shear storage modulus recovered to its initial value. In order to resemble the cytoplasm of a living cell the lysate should be supplied with ATP; otherwise, its viscoelastic properties are changed dramatically. These results clearly indicate that in living cells and in lysates the structure of the fluid changes in the presence of ATP. The rate of ATP consumption, however, is far much lower than in living cells. At this point, the discussion remains open. In the system, some active processes take place and change the structure from high modulus gel to low modulus gel. For example, in the process of stepping kinesin-1, the energy of ATP hydrolysis is used to detach the motor domain (Clancy et al. 2011; Sozanski et al. 2015) from the microtubules. In general, the ATP molecules unglue different components of lysate, change the viscoelastic gel-like structure, and decrease the shear storage modulus of the lysate.

Author contributions AW: rheology measurements, data analysis, data interpretation, manuscript writing; TK: proposing the studies, data interpretation, manuscript writing, figure preparation; JSz: cell culturing, cell lysis; KK: cell culturing, DNA cleavage preparation, data interpretation; KM: data interpretation, manuscript writing; RH: proposing the studies, data interpretation, manuscript writing. All authors reviewed the manuscript. Authors claim no competing financial interests.

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References

- Arevalo RC, Urbach JS, Blair DL (2010) Size-dependent rheology of type-I collagen networks. *Biophys J* 99(8):L65–L67
- Ballerini M, Cabibbo N, Candelier R, Cavagna A, Cisbani E, Giardina I, Lecomte V, Orlandi A, Parisi G, Procaccini A, Viale M, Zdravkovic V (2008) Interaction ruling animal collective behavior depends on topological rather than metric distance: evidence from a field study. *Proc Natl Acad Sci* 105(4):1232–1237
- Ciccolini L, Shamlou PA, Titchener-Hooker N, Ward J, Dunnill P (1998) Time course of sds-alkaline lysis of recombinant bacterial cells for plasmid release. *Biotechnol Bioeng* 60(6):768–770
- Ciccolini L, Shamlou PA, Titchener-Hooker N, Ward J, Dunnill P (1999) Rheological properties of chromosomal and plasmid dna during alkaline lysis reaction. *Bioprocess Eng* 21(3):231–237
- Clancy BE, Behnke-Parks WM, Andreasson JO, Rosenfeld SS, Block SM (2011) A universal pathway for kinesin stepping. *Nat Struct Mol Biol* 18(9):1020–1027
- Dombrowski C, Cisneros L, Chatkaew S, Goldstein RE, Kessler JO (2004) Self-concentration and large-scale coherence in bacterial dynamics. *Phys Rev Lett* 93(9):098103
- Ewoldt RH, Hosoi A, McKinley GH (2008) New measures for characterizing nonlinear viscoelasticity in large amplitude oscillatory shear. *J Rheol* (1978-present) 52(6):1427–1458
- Foffano G, Lintuvuori J, Morozov A, Stratford K, Cates M, Marenduzzo D (2012) Bulk rheology and microrheology of active fluids. *Eur Phys J E* 35(10):1–11
- Kemkemer R, Kling D, Kaufmann D, Gruler H (2000) Elastic properties of nematoid arrangements formed by amoeboid cells. *Eur Phys J E* 1(2-3):215–225
- Kinloch IA, Roberts SA, Windle AH (2002) A rheological study of concentrated aqueous nanotube dispersions. *Polymer* 43(26):7483–7491
- Kong S, Day AF, O’Kennedy RD, Shamlou PA, Titchener-Hooker NJ (2009) Using viscosity-time plots of *Escherichia coli* cells undergoing chemical lysis to measure the impact of physiological changes occurring during batch cell growth. *J Chem Technol Biot* 84(5):696–701
- Levy M, Ciccolini L, Yim S, Tsai J, Titchener-Hooker N, Shamlou PA, Dunnill P (1999) The effects of material properties and fluid flow intensity on plasmid dna recovery during cell lysis. *Chem Eng Sci* 54(15):3171–3178
- Lin YC, Koenderink GH, MacKintosh FC, Weitz DA (2007) Viscoelastic properties of microtubule networks. *Macromolecules* 40(21):7714–7720
- Liu Z, Zhang K, Cheng X (2019) Rheology of bacterial suspensions under confinement. *Rheol Acta* 58(8):439–451
- Malik M, Zhao X, Drlica K (2006) Lethal fragmentation of bacterial chromosomes mediated by dna gyrase and quinolones. *Mol Microbiol* 61(3):810–825
- Marchetti MC, Joanny JF, Ramaswamy S, Liverpool TB, Prost J, Rao M, Simha RA (2013) Hydrodynamics of soft active matter. *Rev Mod Phys* 85(3):1143
- Martinez VA, Clément E, Arlt J, Douarche C, Dawson A, Schwarz-Linek J, Creppy AK, Škultéty V, Morozov AN, Auradou H, et al. (2020) A combined rheometry and imaging study of viscosity reduction in bacterial suspensions. *Proc Natl Acad Sci* 117(5):2326–2331
- Milo R, Phillips R (2015) *Cell biology by the numbers*. Garland Science, New York
- Newton JM, Vlahopoulou J, Zhou Y (2017) Investigating and modelling the effects of cell lysis on the rheological properties of fermentation broths. *Biochem Eng J* 121:38–48
- Palacci J, Abécassis B, Cottin-Bizonne C, Ybert C, Bocquet L (2010) Colloidal motility and pattern formation under rectified diffusiophoresis. *Phys Rev Lett* 104(13):138302
- Parrish JK, Hamner WM (1997) *Animal groups in three dimensions: how species aggregate*. Cambridge University Press, Cambridge
- Parry BR, Surovtsev IV, Cabeen MT, O’Hern CS, Dufresne ER, Jacobs-Wagner C (2014) The bacterial cytoplasm has glass-like properties and is fluidized by metabolic activity. *Cell* 156(1):183–194
- Paxton WF, Kistler KC, Olmeda CC, Sen A, St Angelo SK, Cao Y, Mallouk TE, Lammert PE, Crespi VH (2004) Catalytic nanomotors: autonomous movement of striped nanorods. *J Am Chem Soc* 126(41):13424–13431
- Pelletier V, Gal N, Fournier P, Kilfoil ML (2009) Microrheology of microtubule solutions and actin-microtubule composite networks. *Phys Rev Lett* 102(18):188303
- Sato M, Schwartz WH, Selden SC, Pollard TD (1988) Mechanical properties of brain tubulin and microtubules. *J Cell Biol* 106(4):1205–1211
- Schweitzer F (2007) *Brownian agents and active particles: collective dynamics in the natural and social sciences*. Springer, Berlin
- Serra-Picamal X, Conte V, Vincent R, Anon E, Tambe DT, Bazellieres E, Butler JP, Fredberg JJ, Trepas X (2012) Mechanical waves during tissue expansion. *Nat Phys* 8(8):628
- Sozanski K, Ruhnnow F, Wisniewska A, Tabaka M, Diez S, Holyst R (2015) Small crowders slow down kinesin-1 stepping by hindering motor domain diffusion. *Phys Rev Lett* 115(21):218102
- Stephenson D, Norman F, Cumming R (1991) Shear thickening of dna in sds lysates. *Bioseparation* 3(5):285–289
- Weber SC, Spakowitz AJ, Theriot JA (2010) Bacterial chromosomal loci move subdiffusively through a viscoelastic cytoplasm. *Phys Rev Lett* 104:238102–238102
- Wright JL, Jordan M, Wurm FM (2001) Extraction of plasmid dna using reactor scale alkaline lysis and selective precipitation for scalable transient transfection. *Cytotechnology* 35(3):165–173

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