Published in final edited form as:

Laser Phys. 2014 June 1; 24(6): 065601–.

Measurement of the microscopic viscosities of microfluids with a dynamic optical tweezers system

Yuquan Zhang^{1,5}, Xiaojing Wu^{2,5}, Yijia Wang¹, Siwei Zhu², Bruce Z Gao³, and X-C Yuan⁴

¹Institute of Modern Optics, Key Laboratory of Optoelectronic Information Science and Technology, Ministry of Education of China, Nankai University, Tianjin 300071, People's Republic of China

²Nankai University Affiliated Hospital, Tianjin 300121, People's Republic of China

³Department of Bioengineering, Clemson University, Clemson, SC 29634, USA

⁴Institute of Micro & Nano Optics, Shenzhen University, Shenzhen 518060, People's Republic of China

Abstract

Viscosity coefficients of microfluids—Newtonian and non-Newtonian—were explored through the rotational motion of a particle trapped by optical tweezers in a microflute. Unlike conventional methods based on viscometers, our microfluidic system employs samples of less than 30 μ l to complete a measurement. Viscosity coefficients of ethanol and fetal bovine serum, as typical examples of Newtonian and non-Newtonian fluids, were obtained experimentally, and found to be in excellent agreement with theoretical predictions. Additionally, a practical application to a DNA solution with incremental ethidium bromide content was employed and the results are consistent with clinical data, indicating that our system provides a potentially important complementary tool for use in such biological and medical applications.

Keywords

viscosity measurement; dynamic optical tweezers; microfluid

1. Introduction

Viscosity is an important mechanical property of fluids, revealing the resistance of a fluid that is being deformed by either shear stress or extensional stress. Depending on the relationship between the shear stress and the rate of strain and its derivatives, fluids can be characterized as belonging to the following groups: the Newtonian fluids, whose stress is directly proportional to the rate of strain, with the proportionality, namely the viscosity, remaining constant; the non-Newtonian fluids, where stress is not proportional to the rate of strain, meaning that the viscosity does not remain constant with increase in the flow rate [1, 2]. The ability to gather data on a fluid's rheological characteristics provides researchers

siweiz@medmail.com.cn and xcyuan@nankai.edu.cn.

⁵The authors contributed equally to this paper.

with an important parameter. The interrelation between the rheology and other product dimensions often makes the measurement of viscosity the most sensitive or convenient way of detecting changes in color, density, stability, solids content, and molecular weight [3]. Viscosity measurement plays a very important role in the petroleum, chemical, national defense and medical industries [4]. Measurement of this hydrodynamic property of a fluid can provide high-resolution information about its biological and physical properties, and the information can be useful in modeling any interactions accordingly.

The traditional methods of viscosity measurement are the capillary tube method [5], rotation method [6], and vibration measurement method [7]. Different techniques have been employed in order to characterize the property in recent years: the atomic force microscope (AFM) [8] and ultrasonic techniques [9] were introduced in such applications. Meanwhile, the development of optical technology has promoted the application of viscosity measurement [10–16], in areas such as the fluorescence technique [11], acousto-optical methods [12], and the optical tweezers technique [13–16]. The optical tweezers technique was invented by Ashkin two decades ago [17]; it has been proved to be a powerful tool in biological research [18–22]. Besides numerous applications in life science and physics, it has increasingly been used in engineering for micromanipulation and microassembly [23–28].

In this paper, we demonstrate the technique of using micron sized polymethyl methacrylate (PMMA) particles trapped using dynamic optical tweezers as a probe to measure the local viscosity of microfluidics. As a major difference from the situation for conventional methods requiring a characteristic megadose (at least several milliliters), here we only need a microfluidic sample of less than 30 μl in volume for each measurement in our system. We also expand on the basic theoretical and experimental aspects of this method, such as the dynamic light scattering and gradient forces, as well as velocity measurements. Due to the local absorption of laser radiation within the particles, we employ the conventional optical trapping force to manipulate the microparticles. We calibrate the system by using pure water first, and then measure the viscosities of ethanol (as a typical example of a Newtonian fluid) and fetal bovine serum (FBS; as a typical example of a non-Newtonian fluid) at different velocities. The results show excellent agreement with the reference value. As a simple and effective method, it could have great prospects in medical screening. To corroborate this concept, we also successfully utilize the proposed system to measure the viscosity of DNA solutions doped with different dosages of ethidium bromide (EB) at the end; the results corroborate the high performance of our system.

2. The experimental system and methods

2.1. The optical setup and procedure

Figure 1 shows a schematic of the galvanometer-based optical manipulation system for continuous generation of dynamic steering traps around the optical axis. Mirror galvanometer systems (Cambridge Technology Inc.) are used as beam positioning and beam steering elements in the laser scanning systems. They are typically used with a closed loop servocontrol system. It is designed for beam steering applications with frequency responses of 3–10 kHz. They typically control X and Y directions for laser beams to control the

position of the incident laser spot. With the combination of two independent galvanometer systems, the laser beam can scan along any preselected path. In our experiment, the laser beam was reflected by the galvanometer system to form a circular motion, where the rotating speed was controlled accurately by the servosystem (MiniSAX II servocontroller). The trapping beam is passed through a conjugated lens pair to pinch the beam size, and then enters into the galvanometer system, generating a dynamic circular movement, before being incident on the microscopic objective ($100\times/1.25$, oil, Nikon); thus the trapped particle would follow the same route, just with a demagnified radius.

The laser used for the optical micromanipulation experiment is a 10 W Yb fiber laser (IPG laser GmbH) at 1064 nm. The PMMA particles (Bangs Laboratories Inc.) with the diameter of 3.20 μm are doped into the liquid sample. Experiments were completed in a microflute (figure 2 (a)), employing samples of less than 30 μl for each measurement. The diameter of the sample cell was 2 mm, and the channels had dimensions of 15 mm length, 1 mm width, and 100 μm depth. Inlet and outlet holes (0.320 mm inner diameter) were opened with a punch. Fluid was pumped into the channel by means of a syringe and a syringe pump. Figure 2(b) shows screenshots of a trapped particle rotating along a circumferential trace.

2.2. Trapping force characterization

As the diameter of the PMMA particle is three times the wavelength, Mie scattering theory is used for approximate calculations. The force exerted on the particles can be decomposed into two components: the gradient force and the scattering force. The scattering force always acts along the direction of light propagation, and the gradient force has axial and radial components. The two components balance around the beam focus where the particle is stably trapped: the gravity, the buoyant force, the axial component of the gradient force, and the scattering force are canceled out under equilibrium conditions in the vertical direction; and the radial component of the gradient force, and the drag force are defined in the horizontal direction in the transverse plane. In a single-beam gradient optical trap, the gradient force pulls the trapped particle towards the focus, so the particle is driven dynamically by the beam in the transverse plane. The trapping force is determined by [29]

$$F_{\text{trap}} = \frac{nQP}{c} \quad (1)$$

where P is the trapping laser power, n the refractive index of the medium, c the light velocity, and Q the optical trapping efficiency which is related to the force on the particle and described in terms of a dimensionless parameter.

In a viscous medium, the rotating particle will carry a drag force that opposes its motion. In fluid mechanics, the behavior of fluid flow is primarily characterized by the Reynolds number (Re), which gives a measure of the ratio of inertial forces to viscous forces and consequently quantifies the relative importance of these two kinds of forces for the given flow conditions [30]:

$$Re = \frac{\rho UL}{\mu} \quad (2)$$

where ρ is the fluid's density, U and L denote the characteristic velocity and length (diameter for spheres), and μ is the dynamic viscosity. In microfluidics, characterized by a small size and slow velocity, Re is usually small. In a regime with low Reynolds number, fluid flow is dominated by viscous and surface forces, and the flow field is predictable, stable, and even reversible.

In this work, given the density and dynamic viscosity of water ($0.997 \times 10^3 \text{ kg m}^{-3}$ and $0.890 \times 10^{-3} \text{ N s m}^{-2}$, respectively) at 25°C , the diameter of a trapped PMMA particle of $3.20 \times 10^{-6} \text{ m}$ (Bangs Laboratories, Inc.), and average velocity of less than $2.0 \times 10^{-4} \text{ ms}^{-1}$, Re is calculated to be smaller than 7.169×10^{-4} , which is much lower than 1. Therefore, for the special case of small spherical particles moving slowly through a viscous fluid (and thus at small Re), we can assume the flow to be within the Stokes flow limit, which was derived by George Gabriel Stokes, for the drag force:

$$F_{\text{drag}} = 6\pi\eta av \quad (3)$$

where η is the viscosity of the medium, a is the radius of the particle, and v is the rotating velocity.

On the basis of equation (3), the magnitude of the drag force is directly proportional to the rotating speed. The rotating particle will reach a critical escape velocity when the drag force is equal to the optical trapping force imposed. When such a velocity is reached, the particle falls out of the optical trap but still maintains a broken circular motion at a non-uniform angular velocity. The escape force at the critical escape velocity is approximately evaluated at the state of equilibrium between the drag force and the trap force, i.e.,

$$F_{\text{escape}} = 6\pi a \eta v_{\text{critical}} = F_{\text{trap}} = \frac{nQP}{c}. \quad (4)$$

Consequently, we can rearrange equation (4) and write

$$\eta = \frac{nQP}{6\pi a c v}. \quad (5)$$

The viscosity coefficient is dependent on the trapped particle's size, the refractive index of the surrounding medium, the trapping power, the trapping efficiency, and the rotating velocity.

3. Experimental results and discussion

In this work, the diameter of the trapped particle and the refractive index of the liquid sample are fixed constants. The trapping efficiency Q is a calculated parameter; however, it can be considered constant within the range of permitted error at any trapping power when the setup is stable [31]. Temperature is a crucial factor for viscosity measurement, and a liquid's viscosity tends to decrease as the temperature increases in most circumstances. Hence, the ambient temperature is kept at $25 \pm 0.2^\circ\text{C}$ to confirm the accuracy of the results. Escape velocities were controlled by software (EzCad2, JCZ Technology Co. Ltd), and the mean value of the 'scores' of particles was used to eliminate individual differences. Pure

water is chosen for the calibration of our experimental setup, with its refractive index of 1.326 04 at the wavelength of 1064 nm and viscosity coefficient of 0.890×10^{-3} Pa s (at 25 °C) [32]. Ultimately, Q is calculated as 0.102. Additionally, the particle's diameter is much smaller than the rotating radius, and the velocity is slow enough to allow us to ignore the vortex induced by the revolving particle.

Ethanol has the simplest viscous behavior; it can be treated as a critical Newtonian fluid, because it continues to exemplify fluid properties no matter how quickly it is stirred or mixed. Measured viscosities are shown in table 1; there is a small fluctuation with increasing velocities, a slight deviation appearing as compared with the standard value of 1.074×10^{-3} Pa s at 25 °C [32], which was in accordance with the theory. Each value in the table was calculated from dozens of measurements, and the standard deviation is also shown. In order to verify its accuracy, statistical testing of the hypothesis (a t -test) was carried out for these results. The result indicated that there was no significant difference as compared with the outcome from the standard method at a significance level of 5%, demonstrating the rationality of the proposed method for Newtonian fluids.

Serum is the portion of plasma remaining after the coagulation of blood, including all proteins not used in blood clotting and all the electrolytes, antibodies, antigens, hormones, and any exogenous substances. Due to its very low level of antibodies and to it containing many growth factors, thus allowing for versatility in many different cell culture applications, FBS is the most widely used serum supplement for the *in vitro* cell culture of eukaryotic cells. With these attractive characteristics, FBS also exhibits non-Newtonian viscoelastic fluid dynamics, as shown in table 2. Strikingly different from the case for ethanol, the data analyzed by regression analysis indicated that there was an inverse proportion (linearity) with respect to velocity ($p < 0.01$), which was in correspondence with clinical outcomes.

Compared with traditional methods, our system avoided trivial procedures of cleaning, drying, and other steps. The results demonstrate the accuracy and effectiveness, in the range of permitted errors, of incorporating dynamic optical tweezers in viscosity measurements for both Newtonian and non-Newtonian microfluids, rather than this technique being appropriate for only one category. As regards ethanol, there is a relatively large standard deviation caused by its volatile characteristics, which could be reduced by sealing the sample slot.

4. Potential application in medical screening

Screening in medicine is a kind of medical procedure performed to detect, diagnose, or monitor diseases, and determine a course of treatment. Hydrodynamic measurements that are sensitive to length change (e.g. viscosity) are regarded as the least ambiguous and most critical tests of binding in solution in the absence of crystallographic structural data. DNA is the primary intracellular target of anticancer drugs, and many chemicals exert their antitumor effects through binding to DNA, thereby interrupting the replication of DNA and inhibiting the growth of the tumor cell, which is the basis for designing new and more efficient antitumor drugs, and their effectiveness depends on the mode and affinity of the binding. A classical intercalation model results in lengthening of the DNA helix as base

pairs are separated to accommodate the binding ligand, leading to increase of the DNA viscosity [33, 34]. So, our system is of current interest in view of its potential applications in DNA medical screening.

In order to test the feasibility, competitive ethidium bromide (EB) binding studies were undertaken to gain support for the above concept. EB is well known for its property of intercalating double-stranded DNA (i.e., it inserts itself between the strands), deforming the DNA [35]. It binds with DNA and slips in between its hydrophobic base pairs, and stretches the DNA fragment, removing water molecules from the ethidium cation. This could affect DNA biological processes, like DNA replication and transcription. The result of this dehydrogenation is an increase in the fluorescence of the ethidium, as well as the viscosity.

The original concentration of herring sperm DNA (Promega Corporation, D1815) is 10 mg ml⁻¹, and 100 µl of this solution (i.e., 1 mg DNA) is distributed for each set of samples; this is followed by injecting graded EB into each sample and diluting to 2 ml using normal saline at the end. Consequently, each sample has the same concentration of DNA, but different quantities of EB. To eliminate the internal influence of EB for the solution, we measured the EB solution's viscosity with the concentration given above, while getting rid of the DNA. The results indicate that the viscosities are approximately equal to that of normal saline, leading to the consequence that the internal influence of EB can be ignored in this work. The viscosity experiments were conducted with the temperature still maintained at 25 °C, and figure 3 depicts the measured viscosity of DNA solutions with increasing amounts of EB.

The viscosity of DNA increases with increase in EB content, which is consistent with the observed trend. The accuracy of our system, as shown in figure 3, is high enough for distinguishing the minute oscillations in viscosity relating to R , even from 0 to 0.001. Hence, it could have profound significance in medical screening.

5. Conclusion

In conclusion, we presented a method for measuring the viscosity of a fluid at the microscopic scale based on an efficient dynamic optical tweezers approach. The combination of periodic forces on trapped particles from a highly focused laser beam with microscopic detection leads to a large measurement, which facilitates fast detection in real time. Since the measurement is not based on time and distance, it requires no great accuracy of position calibration and it is simpler to implement using a CCD camera. We have developed a model to relate the escape velocity to the medium viscosity, and demonstrated experimentally that the proposed system was efficient for both Newtonian and non-Newtonian fluids, by measuring the viscosity of ethanol and FBS. The results show excellent agreement between the experiment and the theoretical predictions. Experiments on DNA solutions indicate great accuracy of our system. Comparisons to existing methods show that our optical setup and detection technique allow microscale detection capabilities, which are usually lacking in other techniques. We believe that these qualities of our device could make it a potentially important complementary tool in biological and medical applications that demand precision and high spatial and temporal resolution of viscosity distributions.

Acknowledgments

This work was partially supported by the National Natural Science Foundation of China under grant nos 61036013, 61138003 and 61377052, and the Tianjin Municipal Science and Technology Commission under grant no. 11JCZDJC15200.

References

1. Fox, RW.; McDonald, AT.; Pritchard, PJ. Introduction to Fluid Mechanics. 6th. New York: Wiley; 2003. p. 26-31.
2. Kirby, BJ. Microand Nanoscale Fluid Mechanics: Transport in Microfluidic Devices. Cambridge: Cambridge University Press; 2010. p. 17-31.
3. Steffe, JF. Rheological Methods in Food Process Engineering. 2nd. San Francisco, CA: Freeman; 1996. chapter 1
4. Lane JL, Henderson KO. ASTM Stand. News. 2004; 6:42–45.
5. Wagner RH, Russell J. Anal. Chem. 1948; 20:151.
6. Palmer, AA. US Patent Specification. 4045999. 1977.
7. Primachenko VV. Refract. Indust. Ceram. 1994; 35:168.
8. Ahmed N, Nino DF, Moy VT. Rev. Sci. Instrum. 2001; 72:2731.
9. Chien, HT.; Lawrence, WP.; Raptis, AC.; Sheen, SH. US Patent Specification. 5365778. 1994.
10. Tanner LH. J. Phys. E: Sci. Instrum. 1977; 10:1019.
11. Parker WC, Chakraborty N, Vrikkis R, Elliott G, Smith S, Moyer PJ. Opt. Express. 2010; 18:16607. [PubMed: 20721052]
12. Lou C, Xing D. Appl. Phys. Lett. 2010; 96:211102.
13. Bolognesi G, Bianchi S, Di Leonardo R. Opt. Express. 2011; 19:19245. [PubMed: 21996866]
14. Keen S, Yao A, Leach J, Di Leonardo R, Saunter C, Love G, Cooper J, Padgett M. Lab Chip. 2009; 9:2059. [PubMed: 19568675]
15. Pesce G, Sasso A, Fusco S. Rev. Sci. Instrum. 2005; 76:115105.
16. Nemet BA, CroninGolomb M. Appl. Opt. 2003; 42:1820. [PubMed: 12683762]
17. Ashkin A, Dziedzic JM, Bjorkholm JE, Chu S. Opt. Lett. 1986; 11:288. [PubMed: 19730608]
18. Guck J, et al. Biophys. J. 2005; 88:3689. [PubMed: 15722433]
19. Bao G, Suresh S. Nature Mater. 2003; 2:715. [PubMed: 14593396]
20. Korobtsov A, Kotova S, Losevsky N, Mayorova A, Patlan V, Timchenko E, Lysov N, Zarubina E. Laser Phys. 2012; 22:1265.
21. Gu M, Kuriakose S, Gan X. Opt. Express. 2007; 15:1369. [PubMed: 19532367]
22. Mohanty SK, Uppal A, Gupta PK. Biotechnol. Lett. 2004; 26:971. [PubMed: 15269521]
23. ižmár T, Brzobohatý O, Dholakia K, Zemánek P. Laser Phys. Lett. 2011; 8:50.
24. Hao X, Kuang CF, Li YH, Liu X. Laser Phys. Lett. 2013; 10:045602.
25. Tanaka Y, Kawada H, Hirano K, Ishikawa M, Kitajima H. Opt. Express. 2008; 16:15115. [PubMed: 18795050]
26. Ozkan M, Wang M, Ozkan C, Flynn R, Birkbeck A, Esener S. Biomed. Microdevices. 2003; 5:61.
27. Korda P, Spalding GC, Dufresne ER, Grier DG. Rev. Sci. Instrum. 2003; 73:1956.
28. Roichman Y, Grier DG. Opt. Express. 2005; 13:5434. [PubMed: 19498538]
29. Ashkin A. Biophys. J. 1992; 61:569. [PubMed: 19431818]
30. Happel, J.; Brenner, H. Low Reynolds Number Hydrodynamics: with Special Applications to Particulate Media. Berlin: Springer; 1983. chapter 1
31. Lin C-L, Lee Y-H, Lin C-T, Liu Y-J, Hwang J-L, Chung T-T, Baldeck PL. Opt. Express. 2011; 19:20604. [PubMed: 21997068]
32. Lide, DR. Handbook of Chemistry and Physics. 85th. Boca Raton, FL: CRC; 2004. p. 186-187.
33. Li Y, Yang Z. Inorg. Chim. Acta. 2009; 362:4823.
34. Rao R, Patra AK, Chetana PR. Polyhedron. 2007; 26:5331.

35. Waring MJ. J. Mol. Biol. 1965; 13:269. [PubMed: 5859041]

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Figure 1.
Optical manipulation system that generates dynamic steering traps.

Figure 2.

(a) Schematic of the microfluidic. (b) Screenshots of the trapped particle rotating along a circumferential trace.

Figure 3.
Experimental dependences of a DNA solution's viscosity on increasing amounts of EB.
'R([EB]/[DNA])' on the horizontal axis means the mass ratio of EB to DNA in the samples.

Table 1

Effects of velocity on the measured mean viscosity of ethanol.

Velocity ($\mu\text{m s}^{-1}$)	Viscosity coefficient (10^{-3} Pa s)	Standard deviation
9.86	1.079	0.0092
26.94	1.080	0.0066
32.80	1.074	0.0075
41.58	1.072	0.0062
50.60	1.074	0.0053
68.60	1.071	0.0030
80.66	1.077	0.0037
98.64	1.071	0.0027
114.58	1.069	0.0082
126.58	1.079	0.0104
142.18	1.077	0.0108

Table 2

Effects of velocity on the measured mean viscosity of FBS.

Velocity ($\mu\text{m s}^{-1}$)	Viscosity coefficient (10^{-3} Pa s)	Standard deviation
9.02	1.171	0.0013
15.02	1.155	0.0009
20.94	1.146	0.0013
27.00	1.135	0.0015
38.82	1.131	0.0016
47.64	1.125	0.0013
53.80	1.122	0.0010
65.52	1.106	0.0007
80.70	1.085	0.0016
101.34	1.028	0.0009
137.98	0.985	0.0017
160.32	0.941	0.0008