Point of care diagnostic of the non-linear rheology of biofluids

Author: Carla Riera Llobet
Facultat de Física, Universitat de Barcelona, Diagonal 645, 08028 Barcelona, Spain.

Advisor: Aurora Hernández-Machado

Abstract: We determine the theoretical model for the characterization of the behaviour of Newtonian and non-Newtonian fluids inside a microchannel with segments of different widths. This is done in order to examine the accuracy of a prototype which consists in microscale electronic detection of a fluid/air front advance in order to disclosure different diseases that can be distinguished by the change in the normal rheological characteristics of blood or plasma. We will also test the accuracy of the device when examining Newtonian fluids.

I. INTRODUCTION

Micro and nano scale fluidics has been a widely studied field over the last forty years and has contributed to the progress of technologies such as drug delivery, biological processing, computation or encryption [1]. The advantages of microfluidic devices are their small size, the use of low amount of sample and their ability to reproduce laminar flow conditions, as well as the low effect produced by gravity in comparison with macroscale systems. However, phenomena such as surface tension and capillary forces become relevant.
One potential application for this kind of technology is its use for medical instruments development and/or improvement. Some serious diseases could be diagnosed by the change in the rheological properties of blood [2], a non-Newtonian fluid which its non-linear behaviour varies mainly by the quantity of red blood cells it contains (parameter characterized by the hematocrit). At this moment, the most used technology for this kind of diagnosis are tests based on macroscale analysis of blood samples which cost a lot of time, sample quantity and money.
The RheoDx team is working on the development of a microscale device capable of getting the same results as the ones given by the macroscale machine but using electronic detection of the fluid front flow in a microchannel, which would give information like the viscosity, the shear rate and other rheological properties of the sample.
The first step for achieving this goal is to create a theoretical model for the behaviour of the fluids inside the microchannel and, once the prototype has been manufactured, test its accuracy by running tests with well characterized Newtonian fluids, such as water, and creating a standard through the characterisation of whole healthy blood. This first parts of the process is where this paper is focused on.

II. EXPERIMENTAL SETUP

The experimental configuration consists of a pump connected to a reservoir that holds the fluid inside and which is also connected to the microchannel by a tube. The pump is controlled through a computer with a graphic interface, and the microchannel, fabricated on polydimethylsiloxane (PDMS) attached on glass by using plasma bonding, is on top of a plate which has electrodes printed on it. In Figure 1 we can see an schematic representation of the set-up.

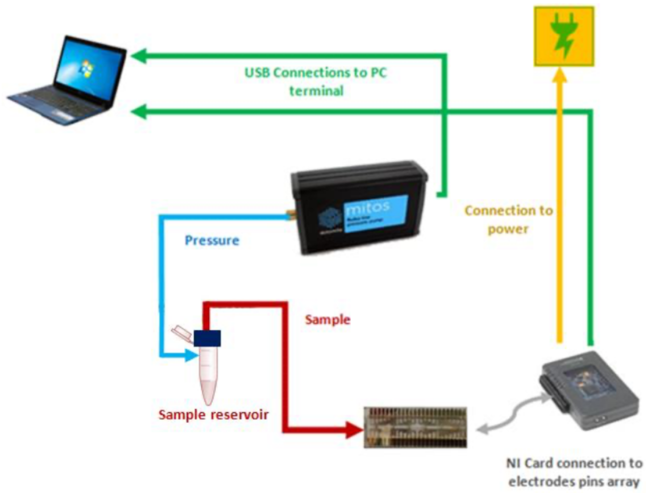

FIG. 1: Experimental setup.

We must remark that in this specific experiment, we have used a microchannel with four different segments which differ in terms of width. This is because the aim of this prototype is to be able to get a complete set of data from the fluid in just one measurement and inducing just one pressure, if we have different widths, we can get a whole profile of data due to geometric factors, while if we just had one width we could only get one single point. Details on the set-up dimensions and the segment widths are included in Table I and Table II respectively.
When we activate the pump, the fluid gets into the microchannel entering by the widest segment, and the front is detected from the connection of an array of electrodes (this is due to the fact that when a fluid with electric conductivity,as water or blood, touches the two paired electrodes these connect, producing an electric signal). The detection is processed by the interface developed by

RheoDx for this prototype, which will return the time the fluid front has spent to reach each pair of electrodes in the microchannel. Then it automatically creates a file .txt type with the time for the 24 pairs of electrodes in the setup.
An easy way to examine the behaviour of the fluid is to study the fluid/air front advancement. In the microchannel, the front velocity $\dot{h}$ is defined as the difference in the fluid front position $h(t)$ through time, and $h(t)$ is defined as the average position of the front in the different fluid layers $h(t) = \frac{1}{N}\sum_{j=1}^{N} h_j(t)$, $h_j(t)$ being the fluid front position in respect to the z axis, as it is shown in Figure 2. This allow us to work with values that are independent of the position in z.
Since we know that between the electrode families there is a gap of $8200\mu m$ and that between the electrodes of the same family is $350\mu m$, with the quick division of

$$\dot{h} = \frac{\Delta d}{\Delta t} \tag{1}$$

where Δd is the separation between the electrodes and Δt the elapsed time, we subtract the mean front velocities of the front for all the sections of the microchannel.

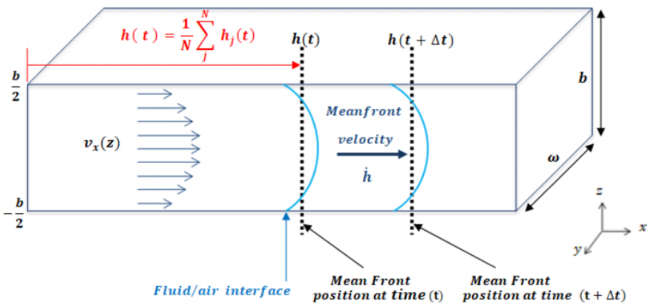

FIG. 2: The velocity $v_x(z)$ is the x component of velocity inside the microchannel on function of the height (z). $h(t)$ is the mean front position of the fluid.

All the values for the different parameters implied in the experiment are

Lenght of the tube l_t (mm)	20
Radius of the tube r (μm)	127
Lenght of the segments l_i (mm)	10
Height of the fluid-air interface in the reservoir h_l (mm)	5
Height of the microchannel b_i (μm)	290

TABLE I: Values of relevance of the experimental setup.

	Segment 1	Segment 2	Segment 3	Segment 4
Segments width w_i (mm)	1	0.85	0.7	0.5

TABLE II: Values of the width of the different channel segments.

III. THEORETICAL MODEL

The simplest characterization model for linear and non-linear fluids would have to be the Power Law Model, also known as the Oswaldt-de-Waele model. In this two-parameter model we relate the stress at which we submit the fluid with the shear rate that at the same time is related to the mean front velocity of the fluid like

$$\dot{\gamma} = \frac{\dot{h}}{b} \tag{2}$$

And so

$$\eta(\dot{\gamma}) = m\dot{\gamma}^{n-1} \tag{3}$$

As we see from this last equation, the viscosity of a non-Newtonian fluids depend on the shear rate as well as of the $(n-1)$ exponent and, is immediate to see, that if $n = 1$, we recover the characteristic linear expression for a Newtonian fluid. In the case of $n < 1$ we would discern Shear Thinning behaviour which means that as shear rate increases there is a descent in viscosity, and if $n > 1$ we would discern Shear Thickening behaviour which is when there is an increase in the viscosity as we increase the shear rate. m would be a parameter that also characterises the nature of the fluid, so if it is Newtonian it will be equal to the viscosity of the fluid while for a non-Newtonian when the $\dot{\gamma} = 1$, m will be the viscosity at that specific value of the shear rate.
We are now going to consider our experimental coupled system. As we have seen before, it is formed by three main components; a reservoir, a tube and a microchannel. We must take into account that the microchannel is divided in four segments of different widths, which will produce pressure drops as we move from one segment to the next, so the pressure difference inside each region will variate. As a result we have

$$\Delta P_i = P_{pump} + \rho g h_l - \Delta P_t - \sum_{j=1}^{i-1} \Delta P_{R_j} - P_{i_{cap}} \tag{4}$$

Where P_{pump} is the pressure exerted by the pump, $\rho g h_l$ is the hydrostatic pressure where h_l is the height of the fluid in the reservoir. ΔP_t is the pressure drop inside the tube

$$\Delta P_t = \frac{2 l_t m \left(\frac{1}{n} + 3\right)^n}{r^{1+n}} {v_t}^n \tag{5}$$

$P_{i_{cap}}$ is the capillary pressure in every segment

$$P_{i_{cap}} = 2\tau cos\theta \left(\frac{1}{b_i} + \frac{1}{w_i}\right) \tag{6}$$

That is calculated by means of the Young-Laplace equation for a rectangular channel and in which τ is the surface tension coefficient between fluid-air, b and w are the height and the width of this and θ is the contact angle of

the meniscus with the walls of the channel, which we have calculated for each segment and exerted pressure using a program in python based on the recognition of circles in an image. It must be calculated for all the different injected pressures.
Lastly ΔP_{R_j} is the resistance pressure due to the filling of every segment of the microchannel before the one where we are measuring

$$\Delta P_{R_j} = \frac{2m}{b_j} l_j \dot{h}_j^n \left[\frac{2}{b_j} \left(\frac{1}{n} + 2 \right) \right]^n \tag{7}$$

The pressure drop inside the tube and inside each microchannel is calculated by means of the Naiver-Stokes equation and the flow rate definition. Here we will display only how it is done for the rectangular microchannel as it is analog for the tube but in that we change to cylindrical coordinates.
We will start by calculating the flow rate of the microchannel. Due to the rectangular geometry $\vec{v} = (v_x(z), 0, 0)$. If we define

$$\dot{\gamma}(z) = \frac{\partial v_x(z)}{\partial z} \tag{8}$$

Now considering $\nabla P = \frac{\Delta P}{h(t)}$ and that the Naiver-Stokes equation when considered a stationary regime is $\nabla \eta \nabla^2 v = \nabla P$ we rewrite

$$\frac{d\eta(\dot{\gamma}(z))\dot{\gamma}(z)}{dz} = \frac{\Delta P}{h(t)} \tag{9}$$

Integrating this we find

$$\dot{\gamma}(z) = \left(\frac{\Delta P}{h(t)m} z \right)^{\frac{1}{n}} \tag{10}$$

To continue we integrate Eq.(8) between $\pm b/2$ with boundary conditions non-slip so we obtain $v_x(z)$

$$v_x(z) = \left(\frac{\Delta P}{h(t)m} \right)^{\frac{1}{n}} \left(\frac{1}{\frac{1}{n}+1} \right) \left(z^{1+\frac{1}{n}} - \left(\frac{b}{2} \right)^{1+\frac{1}{n}} \right) \tag{11}$$

The flow rate

$$Q = 2 \int_0^w dy \int_{-\frac{b}{2}}^{\frac{b}{2}} v_x(z)\, dz = 2w \left(\frac{\Delta P}{h(t)m} \right)^{\frac{1}{n}} \left(\frac{1}{\frac{1}{n}+2} \right) \left(\frac{b}{2} \right)^{2+\frac{1}{n}} \tag{12}$$

It is obvious that in this equations it does not appear the mean front velocity $\dot{h}$ that we get from the experimental data, but knowing the definition of the flow rate $Q = bw\dot{h}(t)$, we can rewrite the general expression for the pressure drop inside the segment as

$$\Delta P(t) = \frac{2m}{b} h(t) \dot{h}(t)^n \left[\frac{2}{b} \left(\frac{1}{n} + 2 \right) \right]^n \tag{13}$$

The resistance pressure will have the same expression but because it is due to the segment being completely filled, $h_i(t)$ and $\dot{h}_i(t)$ are replaced by the length of the segment l_i and the mean front velocity in it $\dot{h}_i$.
Substituting now for each segment using Eq.(4) and taking into account the principle of mass conservation $v_t \pi r^2 = \dot{h}_1 w_1 b_1 = \dot{h}_2 w_2 b_2 = \dot{h}_3 w_3 b_3 = \dot{h}_4 w_4 b_4$, as well as grouping for convenience the terms that are not velocity dependent in a term that we will call effective pressure $\Sigma P_i = P_{pump} + \rho g h_l - P_{i_{cap}}$, we can rewrite a general expression. Also from previously done experiments and results, we know that in the existing conditions of our experimental setup and for all t, the resistance associated to the fluid flow inside the segment in which we are calculating is much smaller than the ones associated to the tube and the previous segments $\Delta P_i(t) << \sum_{j=1}^{i-1} \Delta P_{R_i} + \Delta P_t$. This conditions allow us to have constant velocity in respect to time, $\dot{h} \neq h(t)$.

$$\Sigma P_i = K_i(m, n) \dot{h}_i^n \tag{14}$$

$$K_i(m,n) = m \left[\frac{2l_t \left(\frac{1}{n} + 3 \right)^n}{r^{1+n}} \left(\frac{b_i w_i}{\pi r^2} \right)^n + \sum_{j=1}^{i-1} \frac{l_j \left(\frac{1}{n} + 2 \right)^n}{\left(\frac{b_j}{2} \right)^{1+n}} \left(\frac{w_i b_i}{w_j b_j} \right)^n \right] \tag{15}$$

When $n \neq 1$, this equations will allow us to characterize non-Newtonian fluids, and with $n = 1$ we will recover a model for the Newtonian ones.

IV. EXPERIMENTAL METHOD

As a simple method to determine whether a fluid is Newtonian or Non-Newtonian,we can use a two-parameter power-law expression

$$\Sigma P_i = K_i(m, n) \dot{h}_i^n \tag{16}$$

Where n and m are characterizing fluid constants and specifically n is the one we have already seen in the theoretical model equations. This type of equations are comparable to the ones found with the theoretical model (Eq.(14)).
With the data that we collect from the experiment (the velocity for each applied pressure), we can find equations like Eq.(16) with which we are able to define the fluids nature. But although it would seem like we can control every variable in the ΔP there are factors like fluctuation of the pressure injected by the pump, changes in the contact angle or others that may be significant and which we can not quantify exactly.
In light of this possible effects, we created a little program that will do the following: Firstly it will graph and

show the equations for the Pressure of the pump in front of the velocity but living a free parameter

$$P_{pump} = A_i' \dot{h}_i^{n'} + C_i' \quad (17)$$

C_i' can be interpreted as the rest of the contributions of the exerted effective pressure, so the next step will be to subtract this contribution to the pump pressure and once done that, we will recalculate the equation

$$P_{pump} - C_i' = \Sigma P_{exp_i} = A_i \dot{h}_i^n \quad (18)$$

If we now take logarithms in both sides of Eq.(18) we get

$$\log(\Sigma P_{exp_i}) = \log(A_i) + n \log(\dot{h}_i) \quad (19)$$

The slope of Eq.(19) is n.
If we now compare Eq.(18) to Eq. (14), we can recognize that

$$A_i = K_i(m, n) \quad (20)$$

Isolating m from Eq.(20) we get

$$m = \frac{A_i}{\frac{2l_t\left(\frac{1}{n}+3\right)^n}{r^{1+n}}\left(\frac{b_i w_i}{\pi r^2}\right)^n + \sum_{j=1}^{i-1}\frac{l_j\left(\frac{1}{n}+2\right)^n}{\left(\frac{b_j}{2}\right)^{1+n}}\left(\frac{w_i b_i}{w_j b_j}\right)^n} \quad (21)$$

Recalling Eq.(3), we can rewrite an expression for the viscosity for both Newtonian and Non-Newtonian fluids. For Non-Newtonian we can rewrite a general expression like

$$\eta_i(\dot{\gamma}) = \frac{A_i}{\frac{2l_t\left(\frac{1}{n}+3\right)^n}{r^{1+n}}\left(\frac{b_i w_i}{\pi r^2}\right)^n + \sum_{j=1}^{i-1}\frac{l_j\left(\frac{1}{n}+2\right)^n}{\left(\frac{b_j}{2}\right)^{1+n}}\left(\frac{w_i b_i}{w_j b_j}\right)^n}\dot{\gamma}^{n-1} \quad (22)$$

As for Newtoninan we know that $\eta = m$ so its general expression will be just Eq.(21) but substituting $n = 1$

$$\eta_i = \frac{A_i}{\frac{8l_t}{r^2}\left(\frac{b_i w_i}{\pi r^2}\right) + \sum_{j=1}^{i-1}\frac{12 l_j}{{b_j}^2}\left(\frac{w_i b_i}{w_j b_j}\right)} \quad (23)$$

For each segment the equations will be

$$\eta_1 = \frac{A_1}{\frac{8l_t}{r^2}\left(\frac{b_1 w_1}{\pi r^2}\right)} \quad (24)$$

$$\eta_2 = \frac{A_2}{\frac{8l_t}{r^2}\left(\frac{b_2 w_2}{\pi r^2}\right) + \frac{12 l_1}{{b_1}^2}\left(\frac{w_2 b_2}{w_1 b_1}\right)} \quad (25)$$

$$\eta_3 = \frac{A_3}{\frac{8l_t}{r^2}\left(\frac{b_3 w_3}{\pi r^2}\right) + \frac{12 l_1}{{b_1}^2}\left(\frac{w_3 b_3}{w_1 b_1}\right) + \frac{12 l_2}{{b_2}^2}\left(\frac{w_3 b_3}{w_2 b_2}\right)} \quad (26)$$

$$\eta_4 = \frac{A_4}{\frac{8l_t}{r^2}\left(\frac{b_4 w_4}{\pi r^2}\right) + \frac{12 l_1}{{b_1}^2}\left(\frac{w_4 b_4}{w_1 b_1}\right) + \frac{12 l_2}{{b_2}^2}\left(\frac{w_4 b_4}{w_2 b_2}\right) + \frac{12 l_3}{{b_3}^2}\left(\frac{w_4 b_4}{w_3 b_3}\right)} \quad (27)$$

Due to Newtonian fluids having by definition the viscosity as a characteristic that does not vary for a same fluid, we know that $\eta_1 = \eta_2 = \eta_3 = \eta_4$.
The program will fit the corresponding data to Eq.(18) and Eq.(22) if it is non-Newtonian or to Eq.(21) if it is Newtonian.

V. EXPERIMENTAL RESULTS

We have used water to firstly test our experimental setup accuracy because it is the broadest studied fluid up until now, which leads to its rheological properties to be very precisely characterized. This makes it easy for us to compare our results to them and know with little error if our setup is measuring something that actually makes sense.
For instance we know that water viscosity at 22°C, temperature at which our experiments have been done, is of 1.0016 mPa·s [1] and that, because it is a Newtonian fluid, the value of the n exponent is 1.
From our program we get the following graphs

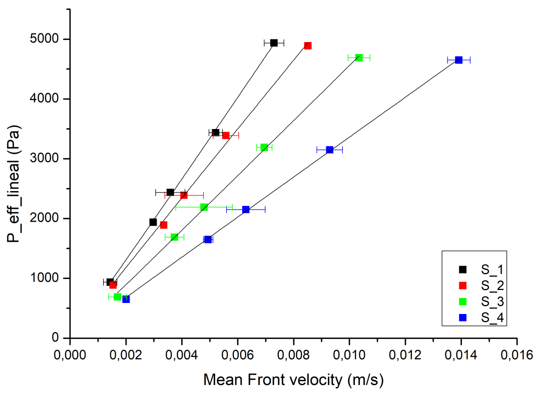

FIG. 3: Effective pressure as a function of velocity for water in the different segments

From Figure 3, where we find the regression of the effective pressure in front of the mean front velocity, we see that for the different segments we get different slopes, this is due to the different geometries of the segments and it is in accordance with our theoretical model.
What is of importance but, it is that we obtain an exponent n which, when we take into account the experimental error, we see how they all fit the value for a Newtonian fluid as water is. The values of n can be found in Table III.

As for Figure 4, and as it is detailed in Table III, the viscosity values are really similar between them and also to the value in the literature [1], which make us consider

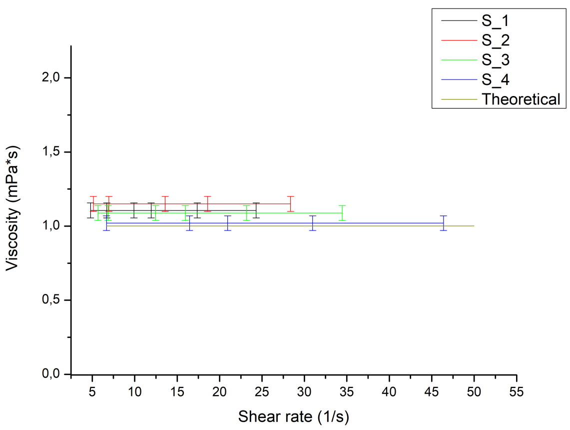

FIG. 4: Viscosity as a function of the shear rate for water in the different segments

that our experimental setup and data processing is adequate to acquire the data we want in an effective way.

	Segment 1	Segment 2	Segment 3	Segment 4
n	1.0228	0.9874	1.0126	0.9929
Viscosity (mPa*s)	1.11±0.05	1.15±0.05	1.08±0.05	1.01±0.05

TABLE III: n exponent and viscosity data of water in each segment of the microchannel.

VI. CONCLUSIONS

We have build a theoretical model to describe both the behaviour of Newtonian and non-Newtonian fluids inside a microchannel with different widths when we apply diverse pressures and we have implemented it as a data analysis program. After conducting experiments with a widely studied Newtonian fluid as water is, we have used this data as the input to our analysis program results from which can be found in the Experimental Results section. From this we may conclude that our model and out experimental device works precisely for the characterization of Newtonian fluids and therefore the next step would be test its performance for Non-Newtonian fluid characterization.

Acknowledgments

I would like to express my most sincere appreciation and gratitude to my advisor Dra.Aurora Hernández-Machado for her continuous support, guidance and her always encouraging point of view. Also to all the members at RheoDx, a group of remarkable people who have always assisted me with great patience and made me feel very comfortable.
Finally I would like to thank my partner, my family and my friends, for without their constant support I do not know if I would have gotten this far or at least doing it while still being sane.

[1] Patrick Tabeling, *Introduction to Microfluidics*, 1st. ed. (Oxford University Press, Oxford, 2005).
[2] Claudia A.Trejo Soto, *Front Microrheology of Biological Fluids*, PH.D Thesis (Universitat de Barcelona,2016).
[3] Marc Pradas-Gené, *Interfaces in Disordered Media*, PH.D Thesis (Universitat de Barcelona,2009).
[4] Trejo-Soto, C., Costa-Miracle, E., Rodriguez-Villarreal, J. Cid, Alarcón, T. i Hernández-Machado, A.. "Capillary Filling at the Microscale: Control of Fluid Front Using Geometry". Plos One **11**: 4-22 (2016).
[5] Trejo-Soto, C., Costa-Miracle, E., Rodriguez-Villarreal, J. Cid, Alarcón, T. i Hernández-Machado, A.. "Front Microrheology of the Non-Newtonian Behaviour of Blood: Scaling Theory of Erythrocyte Aggregation by Aging". Soft Matter **13**: 3042-3047 (2017).
[6] Méndez-Mora, L., Cabello-Fusarés, M., Farré-Torres, J., Riera-Llobet, C., López, S., Trejo-Soto, C., Alarcón-Cor, T. i Hernández-Machado, A.. "Electro Micro-rheometer with biofluids". Preprint (2020)