A Superparamagnetic bead driven fluidic device

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ABSTRACT

Injection strategies have been employed in the field of fluidic MEMS using piezo electric or thermal actuators. A very popular application for such technology is inkjet printing. Largely this technology is used to produce droplets of fluid in air; the aim of this investigation is to produce an injection device for the precise dispensing of nanolitre volumes of fluid. A novel technique for dispensing fluid using superparamagnetic beads has been investigated. The beads used (Dynal Biotech) contain a homogeneous dispersion of Fe_2O_3, allowing for easy control with a magnet. This magnetic property is exploited, by a plug of approximately 60 000 beads within a micro channel. This is accomplished by applying a non-uniform magnetic field from a bullet magnet within close proximity of the bead plug. Once the plug is formed it can be moved along the micro channel by moving the magnet and thus, provide a plunger-like action.

Previous work has demonstrated a bead plug device is able to dispense fluid from a micro channel at rates up to 7.2 $\mu l min^{-1}$. This is an investigation using silicon and Pyrex fabricated micro channels with smaller dimensions, such that the dimensions will be similar to those which will be used to produce a pipette device. Here results are presented using these fabricated micro channels, where the effects of using differently sized bead plugs and varying velocities are examined. The results follow our proposed theory; further analysis is required to determine the operation of a bead plug during all states of movement.

Keywords: Superparamagnetic beads, Micro channel, Micro fluidics, Nano-volume pipette

1. INTRODUCTION

Micro Electro Mechanical Systems (MEMS) for biological applications has seen an increase in the development of micro fluidic components. These components include pumps,[1,2] mixers[3,4] and separators[5] to name a few. Combined these components will be used to form Lab-on-a-Chip systems or Micro Total analysis systems (μTAS).[6,7] Such systems will be used to mimic and where appropriate, replace conventional bench top equipment. The obvious advantage of replacing these larger systems is the reduced sample and reagents volumes required. These reduced volumes are due to the ability to produce very small devices. In addition the reduced dimensions of these MEMS devices require less power and can sometimes produce faster analysis speeds and therefore produce a larger throughput.

A method for the precise metering of samples or reagents is required in small volume micro fluidic systems. This would traditionally be carried out by hand using a pipette, but to produce a complete Lab-on-a-Chip system, this operation needs to be included. Injection strategies for nanolitre volumes have been employed within fluidic MEMS, typically using Piezo electric[8] (PZT) or thermal actuation.[9,10] Such injection strategies are used for inkjet printing and dot spotters where a precise quantity of fluid is dispensed accurately over and over again.

A variety of production techniques have been developed to produce these fluidic MEMS systems. These techniques vary from imprinting using etched shapes to form three dimensional structures in a soft material, to the use of silicon fabrication techniques. Silicon-chip fabrication techniques have been developed over the years to produce electronic structures using a variety of methods, but these methods are now being used to produce MEMS devices. The production of MEMS devices is not dissimilar, using some standard processing, where more

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consideration is sometimes required. A simple channel can be etched in a silicon substrate and capped with Pyrex using anodic bonding. This method allows the channel to be viewed through the Pyrex. Though this is a simple example it could be made more complex by the addition of heating elements in reaction chambers and the addition of diaphragms for pumping actions, a PCR device can be realized.[11]

The results presented here provide details for the creation of a pipette to controllably dispense nanolitre volumes of fluid using magnetic actuation. Such a pipette device system will be used for dispensing variable volumes of different fluids. The fabrication and modelling of such a device is described here and the experimental results discussed.

2. DEVICE CONCEPT

2.1. Injection device

The purpose of this work is to produce a pipette for controllably dispensing a volume of fluid in the low nanolitre range. The device could be integrated with other fluidic devices to form a micro fluidic system. The pipette device could be tailored to inject any fluid as required, but preferably should be able to dispense reagents without heating it above room temperature.

2.2. Magnetic actuation

Magnetic actuation has been used in MEMS structures including fluidic devices. This actuation technique has been implemented using permanent magnets and micro coils to form electro magnets, even a combination of both. This actuation technique has been used to produce linear, revolving, reciprocating motion and separation of magnetic parts or particles. This motion produced using magnetic actuation has been successfully used to produce pipettes,[12,13] micro pumps[14–17] and bio-separators.[5,18]

Magnetic actuation is very versatile and has already been seen as an actuation technique within MEMS devices. It can be simple to implement using just permanent magnets. Magnetic actuation has been chosen for this work due to the simplicity of the technique and there is no contact between the source magnet and the actuator and both could be outside of the fluidic system. This allows for simple devices to be produced which could be cleaned out after use; ready to be used again or disposed of after one use. Due to the simplicity the magnetic actuator can be separate from the fluidic device, allowing for simple fabrication.

Magnetic actuation has already been used to produce pipettes, but this has been carried out using a ferro fluid. An alternative to his technique has been sought after in this work, due to fear of contamination during actuation that a ferro fluid might produce. Ferro fluids are commonly ferrous oxide (Fe_2O_3) suspended in oil, which could mix with the injected fluid and Fe_2O_3 may begin to be removed from the oil suspension during actuation. Finally the technique which has been identified here to produce a pipette device uses superparamagnetic beads with no oil suspension to produce a piston-like plug for pumping. This allows for a greater freedom of manipulation because the beads can be moved through the pumped fluid with no risk of contamination.

2.3. Superparamagnetic Beads

The technique which has been developed to control fluid in a micro channel uses superparamagnetic beads. The beads used for this work were actually produced for a separation technique. The technique involves the surface modification of the beads, such that molecules attach to the surface. The beads are then removed from the solution along with the molecules. This is a quick and easy separation technique. In addition once the beads have been used, they can be washed and used again. Due to the ease of control with a magnet the beads have been chosen for this work[19]

The beads are manufactured by Dynal Biotech, Norway and are available in a variety of sizes and surface modifications. The beads are superparamagnetic monodispersed polymer spheres. Each bead has an even dispersion of magnetic material (Fe_2O_3), encased within a thin polymer shell. Due to this iron content (Fe_2O_3), the beads become dipoles when a magnetic field is applied. This will result in the beads being orientated according to their polarity. When the beads are held by the field of a magnet it will be possible to move them through a channel.

The particular beads used for this technique are epoxy coated, $4.5\mu m$ diameter (M450 Epoxy). The epoxy coating is advantageous for this technique because it makes the beads hydrophobic and in this case non-specific aggregation as well as non-specific adhesion to the micro channel surface. The beads can be introduced in an aqueous solution and consequently trapped in order to be used for this technique. Table 1 summarizes the bead characteristics.

Diameter	$4.5\mu m$
Density	$1.5g/cm^3$
Surface area	$0.9m^2/g$
Iron Content	Approx. 20%
Susceptibilty	0.024

Table 1. Bead characteristics.[19]

2.4. Device operation

The operation of the device proposed during this work is very much like a syringe. This technique uses a piston-like plunger of beads to force fluid from a micro channel. Similar devices have been produced,[12, 13] using a ferro fluid to form a plunger. The beads used for this technique are introduced into a micro channel in a aqueous solution. Once in the micro channel, the beads can be transferred from one fluid to another, providing the beads are not allowed to dry out. If the beads are allowed to dry out, aggregation and adhesion to the surfaces of the micro channel occurs. By moving the beads through the micro channel, fluid is forced out ahead of the piston-plug as happens when a plunger is used in a syringe.

This technique is able to produce a variable dispensing pipette. To introduce the beads, a droplet of water containing the beads is placed at the inlet of the device. The beads are then drawn into the micro channel with a permanent magnet. The beads can then be held in place using a magnet until the injection is required. In order to use the beads to move fluid through the micro channel, first the beads must be clumped together to form a plug. Due to the small dimension of the beads compared to the channel, this plug will effectively block the channel. To achieve this plug the magnet is moved to a position beneath the beads, such that the beads are in the point of highest field gradient of the magnet; which will be at the edge. Once the plug is formed the magnet can be moved along the length of the channel, thus moving the plug and the fluid. The plug will displace a quantity of fluid equal to that of the volume which has been swept out by the plug. By varying the distance the plug is moved the dispensed volume of fluid is varied.

3. THEORY

For this work superparamagnetic beads have been chosen due to the magnetic properties and fluid compatibility of the beads. The beads will orientate much like a dipole when placed within a non-uniform magnetic field. Due to this property the beads posses a magnetic moment, which results in a force which is described by Equation 1. The theoretical force obtained using Equation 1, is for a single bead. For the purposes of this investigation this theoretical force for a single bead is multiplied by the number of beads used to obtain the plug force. In Equation 1 F_B is a single bead force, V_B is the volume of the bead, ΔX is the difference in magnetic susceptibility of the bead (manufacturer data) and the liquid medium, ∇B is the magnetic field gradient, B is the magnetic flux density and μ_0 is the magnetic permeability of a vacuum. The analysis and evaluation of this relationship has already been carried out in previous work by Zborowski.[20] In this case the evaluation was done to analyze the force required for manipulating magnetic beads for cell separation.

$$F_B = \frac{V_B * \Delta X * B * \nabla B}{\mu_0} \tag{1}$$

The force required to move the fluid through the microchannel is determined by the pressure drop which will occur along the length of fluid, due to the fluid flow. The relationship of the force and flow rate is described in Equation 2 and is a form of the well known Haagen-Poiseuille equation. In Equation 2, Q is the flow rate, F_p is the applied plug force, a is the channel width, b is the channel depth, A is the channel area $(a * b)$ and L is the channel length. By substituting Equation 1 into Equation 2 it is possible to determine the maximum flow rate which can be produced for a given bead plug. The flow rate obtained is a volume flow rate, therefore by dividing by the area of the channel (A), the maximum plug velocity can be obtained, Equation 3. If the maximum velocity is exceeded it is obvious that the bead will no longer act like a perfect plunger, but will begin to slip. This slip will result in a lower displacement of fluid for a given plug movement.

$$Q = F_p * \frac{a^3 b^3}{8\mu(a+b)^2 AL} \tag{2}$$

$$Maximum\ Velocity = \frac{F_p * \frac{a^3 b^3}{8\mu(a+b)^2 AL}}{A} \tag{3}$$

4. EXPERIMENTAL

4.1. Purpose of experiment

For this technique to be fully utilized, some experiments have been carried out in order to determine the characteristics of differently sized bead plugs. The experiments were done in order to evaluate how the size of the plug and the speed at which it is moved affects the flow of fluid out of the device. By varying the velocity at which the plug is moved the properties of the bead plug will vary as described in Section 3. The bead plug only exerts a given force dependant on the number of beads and the magnetic field (Equation 1). The size of the force required to displace the water will vary with the flow rate (Equation 2). As the flow rate increases the force required to displace the fluid at this flow rate increases due to the increase in pressure. Once the point at which the plug induced force is no longer large enough, the plug will begin to disperse. This dispersion results in the piston-plug slipping, allowing fluid to pass through and around the plug. It is hoped the results from these experiments will be used to determine a relationship between the plug force and the fluid displaced for varying velocity of the piston-plug. Finally it is hoped this technique can be used to produce a plunger / valve system to both dispense and refill a pipette device.

4.2. Experimental setup

The final pipette device will be produced in silicon and Pyrex, so for the purposes of this experiment, silicon and Pyrex were used to produce a fluidic channel. Single fluidic channels were produced in a silicon substrate using standard fabrication techniques. The channels were DRIE (Deep Reactive Ion Etching) etched in the silicon and capped using Pyrex, with anodic bonding. The channels have been etched such that they are 100 μm wide and 26 μm deep. These dimensions have been used, so it is possible to measure the volume of fluid which is dispensed and it is possible to introduce the beads into the device. The device used is illustrated in Figure 1.

During the experiment it is essential the beads are free to move within the micro channel. To avoid aggregation and beads sticking to the walls of the channel, Triton 1% is introduced into the micro channel at the beginning of the experiment. Once the channel is filled and the fluid is stationary; the beads can be introduced. The beads are introduced in solution at the inlet then gathered using a magnet. The beads now form a plug which can be freely used through the channel.

Due to the small dimensions of the channels used for the experiment it is important to ensure the flow being measured is due to the induced flow from the magnetic beads. To avoid capillary action which will induce

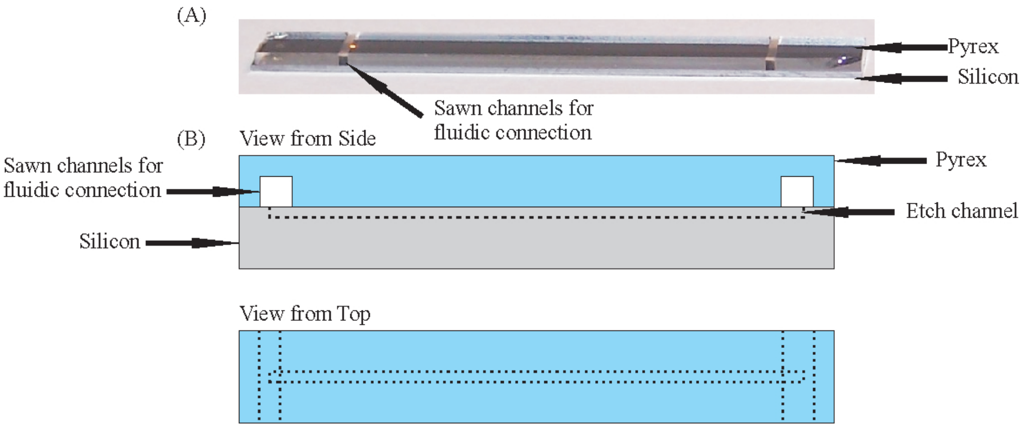

Figure 1. (A) Illustration of the Silicon and Pyrex fabricated micro channel, (B) Image of the silicon and Pyrex micro channel.

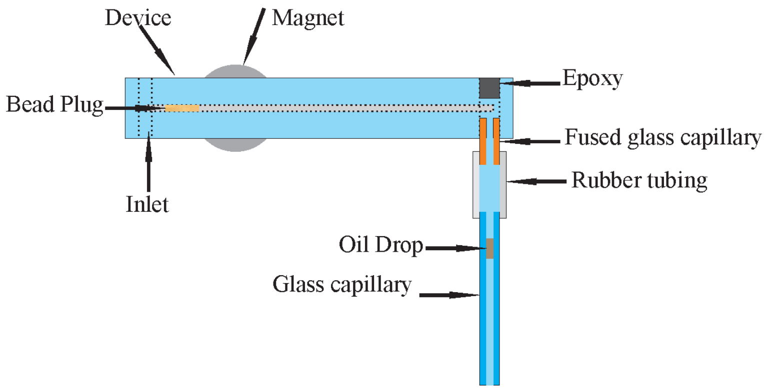

Figure 2. Experimental setup, plan view.

flow within the micro channel, the channel is primed with fluid at the beginning of the experiment. The fluid is introduced into the inlet of the device and allowed to fill the channel using capillary forces. Once filled the fluid is drawn through the remaining system using a syringe to avoid bubble formation. Bubbles will also affect the experiment because the bead plunger could compress the bubbles and not move the fluid. The experimental setup is illustrated in Figure 2.

The micro channel has a fused glass capillary glued at the outlet using epoxy glue. Epoxy is used to block the other part of the outlet. Connected to this capillary is a glass capillary (Cammag, 0.5 μl) connected via a piece of rubber tubing (SF Medical 0.02ID). The glass capillary will be used to measure the flow induced by the bead plug. Finally in order to determine the flow within the micro channel, an oil droplet has been introduced into the glass capillary. The purpose of the oil is to provide a marker to measure flow within the glass capillary.

The magnetic field for these experiments was provided by a two bullet magnets from Assemtech (M1219-5 and M1219-4). These are Neodymium Iron Boron magnets which have a high magnetic field (0.35 $Tesla$, 0.25 $Tesla$), but are only 10 mm and 5 mm in diameter respectively. To ensure the theoretical plug force could be calculated accurately the magnetic flux density of the magnets was measured using a Hall probe. As the beads will be clumped at the edge of the magnet this is the point of most interest. Table 2 summarises measured magnetic flux density and gradient.

During the experiment the magnets are placed beneath and in contact with the channel, such that the centre of the magnet is aligned with the channel, illustrated in Figure 2. The movement for the magnet was provided

Magnet	Magnetic Flux Density(B)	Magnetic gradient(∇B)
M1219-5	0.174 $Tesla$	178 $Teslam^{-1}$
M1219-4	0.125 $Tesla$	83 $Teslam^{-1}$

Table 2. Measured magnetic flux density and gradient of two magnets used for experiments.

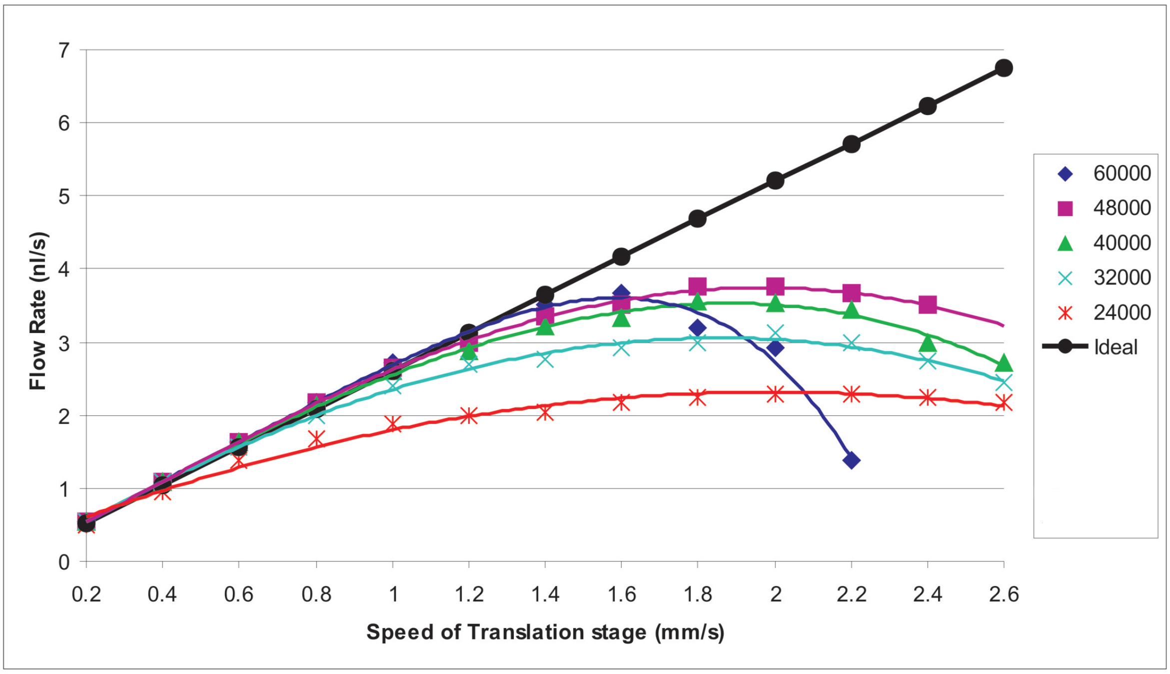

Figure 3. Graph to show flow rate against speed of translation stage for different number of beads for M1219-5 magnet.

by a Melles Griot motorised linear translation stage($NanoStep^{TM}$ 1000 linear positioning stage, with an apt^{TM} system stepper motor controller). The stage is used to move the magnet relative to the micro channel, at varying velocities and distances. For the experiment the stage was used to move the magnet a distance of 10mm and return, at velocities from 0.2 mms^{-1} to 2.6 mms^{-1}. During the experiment the bead plug was allowed to clump to form a plunger before starting the movement for the experiment. Measurements were only taken once the linear translation stage was at a constant velocity. This also applied to the deceleration of the stage and bead plug.

5. RESULTS AND DISCUSSION

The results from the experiments have been displayed in two forms. The first shows the flow rate against bead velocity and the second shows the volume of fluid which is displaced against the bead velocity. The results have been shown in this way because the flow rate shows the rate of the fluid being dispensed, but the volume of fluid displaced is a better method to determine the operation of the pipette.

The results for the flow rate against plug speed are illustrated in Figure 3 for the larger magnet (M1219-5). To better determine the point at which the bead plug no longer acts as a perfect plunger, a set of theoretical data points have been included in this graph, labelled "IDEAL". These "IDEAL" data points represent a bead plug that does not slip at any velocity. It is clear from the results illustrated in this graph that as the velocity at which the plug is increases the flow rate increases. This situation however only occurs until the bead plug begins to slip. This slippage point is when the force provided by the plug can no longer equal the required force to move the fluid for a given velocity. The movement of the plug continues to displace the fluid, but at a lower flow rate due to the slippage past the plug.

The point at which the bead plugs slip is due to the force which can be exerted by the plug, which is directly related to the number of beads; more beads exert a greater force, less beads exert a lower force. The graph clearly illustrates that the point at which the bead plugs slip increases with plug size. However the plug size can only be increased to a certain point. If the plug contains a high number of beads (approximately 60 000), the beads are no longer able to keep up with the magnet, resulting in a dispersion along the channel. This can be clearly seen in the case of the 60 000 bead plug which is able to keep up with the magnet until the point at which it would be expected to slip. After this point the plug begins to disperse and the flow rate decreases dramatically. For all the other size plugs the flow rate falls off slowly, until the point at which the beads are dispersed resulting in the experiment being terminated. This dispersion however is not only a result of the plug force not being great enough, but the magnetic field around the magnet not being great enough. This is because the bead plugs are formed and moved using the edge of the magnet, therefore the plug tends to trail behind the magnet. The combination of a high velocity and a large plug results in the beads being left behind. This situation needs to be avoided, as beads might be lost, resulting in possible contamination of the dispensed fluid. This applies to all the plugs sizes investigated with exception of the 24 000 bead plug. It is believed that this plug is suitably small that the field is surrounding the magnet is large enough to continue moving the plug at velocities beyond those used in this investigation.

The results illustrated in Figure 4 can be used to draw the same conclusions as Figure 3, but also show the volume of fluid which is displaced in a bead translation of 10 mm. It is clear from these results that all of the bead plugs except 24 000, are able to completely plug the channel and dispense a volume of fluid equal to that of the volume which the plug moves through. This is true until the bead plug begins to slip and the volume displaced begins to decrease. In the case of the 24 000 bead plug, slip of fluid past the plug occurs even at low velocities of 0.2 mms^{-1}. From this it would suggest that the 24 000 bead plug is unable to plug the channel at the velocities investigated here. To summarize the results, Table 3 contains the theoretical maximum velocity and actual maximum velocity for each bead plug. The theoretical velocity has been obtained using Equation 3.

Number of beads	**Theoretical maximum**	**Experimental maximum**
60 000	$1.04mms^{-1}$	$1.0mms^{-1}$
48 000	$0.83mms^{-1}$	$0.80mms^{-1}$
40 000	$0.69mms^{-1}$	$0.60mms^{-1}$
32 000	$0.55mms^{-1}$	$0.40mms^{-1}$
24 000	$0.41mms^{-1}$	$0.20mms^{-1}$

Table 3. Theoretical and Experimental magnet velocities for M1219-5 magnet with different plug sizes.

It is clear from the results in Table 3 that the experimental and theoretical values differ. The theoretical value is higher than the experimental value. The theoretical value has been calculated using the properties of the magnets as shown in Table 2. These values will not be the same for all the beads in the plug, because the beads further from the magnet will experience a smaller magnetic flux density and gradient as those closer to the magnet. In addition to the force calculation error, an experimental error while taking measurements to determine the amount of fluid displaced could occur. Finally the error could be due to the number of beads used to form the plug being more or less than expected, if the concentration of the beads in the solution is different than stated. Further investigation will be carried out to better determine the cause of this difference.

The same experiment was repeated with a smaller size magnet (M1219-4) using the same plugs and channel. This produced similar results which are illustrated in Figure 5. It is clear from these results that with the smaller magnet with a lower magnetic flux density that the slip point of the bead is at a lower velocity. Once the bead plug has started to slip the following flow rates are less than that for the larger magnet.

Three of the plug sizes (48 000, 40 000 and 38 000) are similar in operation with slip and continuing opera-

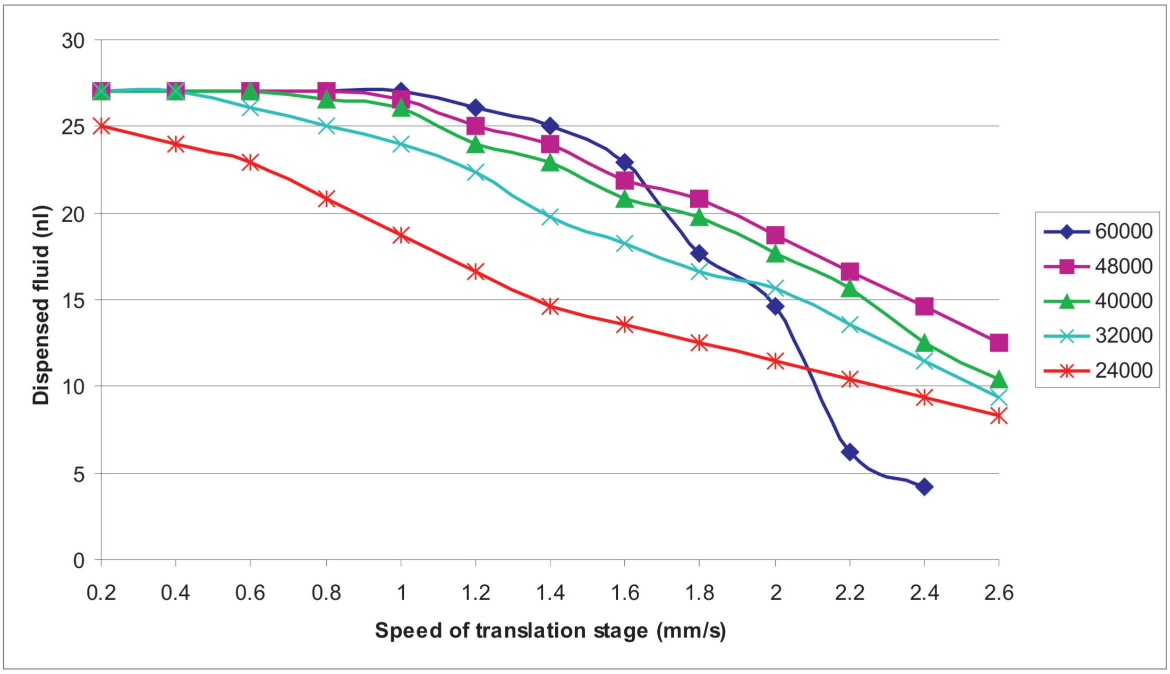

Figure 4. Graph to show fluid dispensed from device against speed of translation stage for different number of beads for M1219-5 magnet.

tion at a less than "IDEAL" flow rate. However the largest plug at 60 000 beads does not move at very high velocity, only 0.6 mm^s (not clearly shown), before it slips. This is a result of the magnet size not being large enough. The smallest plug at 24 000 beads appears to have a similar flow rate against bead magnet velocity as the larger magnet. It is believed this is due to the fact that the beads are always moving through the fluid so the fluid is being dragged with the beads and not displaced with a solid plunger as expected. Therefore for the small number of beads the size of the magnetic field does not appear to affect the flow rate.

6. CONCLUSION

It is clear from the results that a bead plug can be used to displace fluid from a micro channel. The amount of fluid which is displaced from the micro channel can be varied by varying the distance the bead plug is moved and using a magnet velocity below the plug slip velocity. The bead plug can also be moved a fixed distance and the magnet velocity could be varied. Both of these would result in a device able to dispense fluid from a micro channel.

The experiments have focused on obtaining the behaviour of the bead plugs at different velocities but ultimately a pipette device needs to be produced. By utilising the effects of the bead plug slipping at high magnet velocities, a device could be produced which would act as a pipette which is able to refill. This property would have to be combined with a restriction at the outlet of the device. The plug properties and restriction could be used with a straight channel to form a pipette device. When moving the bead plug toward the outlet of the device at a low velocity, such that the bead plug does not slip, fluid will be dispensed at the outlet. As the fluid is dispensed at the outlet, the channel behind the bead plug will be refilled. The plug can be returned back to the starting position at a high velocity, such that the beads will be dispersed, thus moving through the fluid and not inducing movement. If the restriction takes the form of a uni-directional valve, this bead plug technique will be enhanced. The result of this is a simple pipette device, within a straight channel.

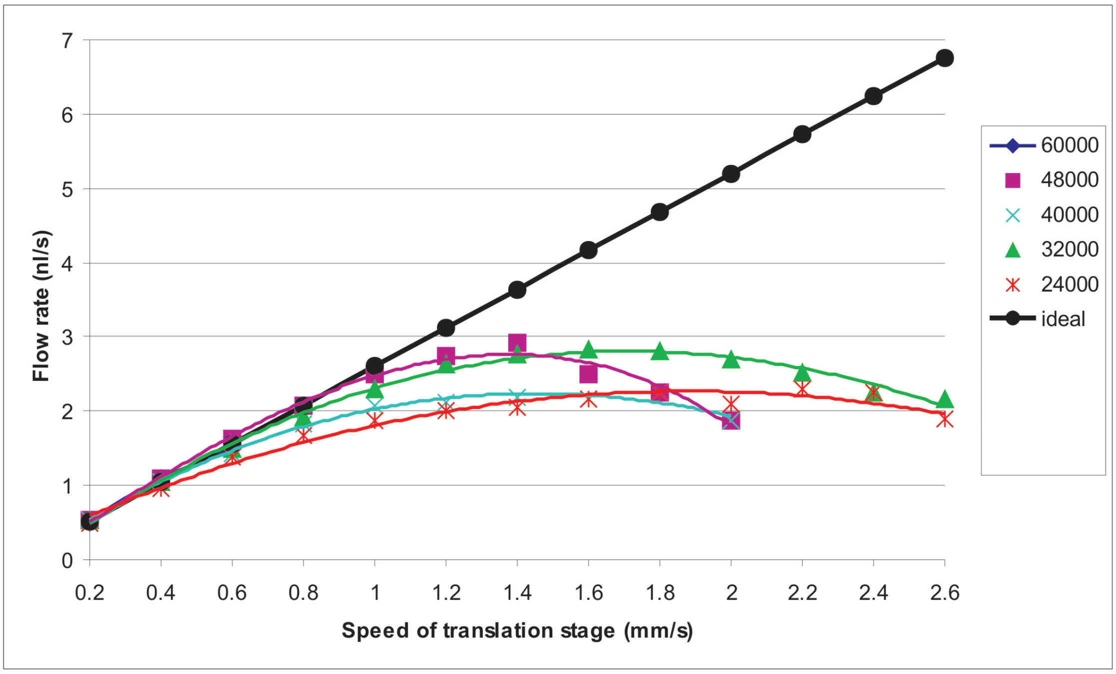

Figure 5. Graph to show fluid dispensed from device against speed of translation stage for different number of beads for M1219-4 magnet.

Though the point of slip can be determined with some degree of error, it is not fully understood why the bead plugs slip in such a way after the plug begins to slip. This requires further analysis of the results to determine whether it is just he beads displacing the fluid or the fluid being dragged by the beads, or a combination of both of these effects.

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