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FLOW SIMULATION OF A NATURAL POLYMER IN A SYRINGE-NEEDLE DELIVERY DEVICE

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Abstract. Neurodegenerative diseases, such as Parkinson's disease, affect a large number of the erderly population and still remain untreated. In recent years, cell therapy has emerged as a promising therapeutic strategy. To increase cell viability, biomaterials are often used as scaffolds and facilitate cell deposition, through injection, to the site of interest. However, fluid forces acting on the cells during injection may lead to their disruption or death. This study aims to develop a novel device for the delivery of a cell-embedded, *in situ* forming, collagen hydrogel. A preliminary simulation study on constricted channels representing the syringe was performed to gain insight into the effect of needle diameter and syringe geometry. Straight needles emanating co-axially from syringes of various geometries were computationally modelled in the two-dimensional space, using OpenFOAM®. The natural collagen solution was modelled as a continuum medium, without cells, and the flow was assumed incompressible, with non-Newtonian fluid constitutive behaviour. The effects of needle diameter and syringe geometry on velocity and shear stresses were examined. The results highlight the importance of geometric characteristics on the design of new cell delivery devices. If cells pass from the syringe barrel to the needle, the pressure drop and the increased velocity could damage them. This is more likely to occur using higher Gauge needles. Further analysis is required including simulations of cells during injection and analysis of their deformation.

1 INTRODUCTION

Parkinson's disease is a challenging neurodegenerative disorder, characterised by dopamine loss, caused by the degeneration of the nigrostriatal dopaminergic system [1]. The cells affected are the dopaminergic neurons in the substantia nigra that send axonal projections to the corpus striatum [2]. Several therapies to address Parkinson's disease have emerged, such as the combination of neurotrophic factors, stem cell therapy, and biomaterials [3].

Despite these recent advancements, clinical translation is still restricted, partially due to limitations in delivering therapeutics to the Central Nervous System (CNS). Since systemic administration cannot reach the CNS due to the limited diffusion of molecules through the blood brain barrier (BBB) [4], an alternative method that offers highly concentrated deposition in the diseased region is the intrastriatal delivery of a cell-embedded, *in situ* forming, collagen hydrogel [5].

Collagen, in recent years, has been widely used in numerous biomedical applications due to its biocompatibility, biodegradability, and weak antigenicity [6]. Therefore, it can offer a protective environment to the cells delivered by preventing the astrogliosis and microgliosis, and by prolonging the cell viability on the site of delivery [7]. Furthermore, collagen is widely used in biomedical applications because of its stability and strength, due to fibre formation caused by self-aggregation and cross-linking [8]. As a natural polymer, collagen has been shown to exhibit a non-Newtonian behaviour, characterised by shear thinning properties [9], that is, its viscosity decreases with increasing shear rate [10].

Even though the strategy of utilising an injectable collagen scaffold to deliver cell therapy to the brain is promising, it still has disadvantages, since the instrastriatal injection required for the delivery can cause tissue damage, neuroimmune response, and haemorrhage. To minimise these responses, an optimised medical device should be designed. Of main consideration is the volume dispensed and the needle dimensions. Current approaches use 18 to 20 Gauge diameter needles and multiple cranial penetrations to achieve a high volume of cell-embedded, *in situ* forming, collagen hydrogel that would allow therapeutic effects. This can cause tissue damage and haemorrhage due to the relatively large diameter of the needle [11]. Thus, it would be beneficial to explore the possibility of using a needle with a smaller diameter than the current standards for intrastriatal injection.

Another important consideration is the geometric design of the syringe used for the intrastriatal injection. As a common practice, low dead space syringes are used in a variety of medical applications since they ensure accurate dosing. Low dead space syringes manage to limit the dead space between the syringe hub and the needle, however this can be achieved with more than one geometric design. Therefore, it is useful to investigate how the design of the syringe could affect the delivery of the cell-embedded collagen.

Finally, in the case of cell therapy, it is highly important to ensure a prolonged viability of the delivered cells, in order to allow them to proliferate and express their therapeutic effects. Although this is partially achieved by the collagen hydrogel, which protects the cells from immune responses once in the site of delivery, their viability must also be ensured during their passage through the delivery device. When cells flow through a syringe needle, forces applied by the fluid on the cells could lead to cell disruption and death [12]. These forces include pressure drop across the cell, shearing forces due to shear flow, and stretching forces due to extensional flow [13].

This study aims to develop a novel device for the effective intrastriatal delivery of a cell-embedded, *in situ* forming, collagen hydrogel to the CNS. A preliminary simulation study on constricted channels representing the needle was performed, and is presented here, with the scope to gain insight into the optimal needle diameter and the effect of the syringe geometry on collagen flow.

2 METHODS

2.1 Collagen shear thinning behaviour

The shear thinning behaviour of soluble collagen, which could be used as scaffold for therapeutic cell delivery, was characterised based on measurements of viscosity, with a rotational viscometer, during controlled increase of the shear rate. For that purpose, pepsin-extracted soluble collagen was used at 3 mg/ml (supplied by Collagen Solutions Plc) and was further diluted to 2 mg/ml, using 10 mM HCl. The temperature on the rotational viscometer was kept stable at 22^oC by a circulating water bath. The rotations per minute (rpm) were gradually increased and the apparent viscosity was measured. Measurements were considered valid when the torque was higher than 12.5%.

The results were analysed by using the well-known power law model [14], which is used to approximate the behaviour of shear-thinning and shear-thickening non-Newtonian fluids and is described by the equation:

$$\eta = K \cdot \gamma^{n-1} \tag{1}$$

where η is the viscosity, γ is the shear rate, K is the flow consistency index and n is the flow behaviour index. The following logarithmic equation:

$$log(\eta) = log(K) + (n-1) \cdot log(\gamma) \tag{2}$$

is used to extract the flow consistency index and the flow behaviour index. n is indicative of the behaviour of the fluid; for $n = 1$, Newtonian behaviour is expected, for $n < 1$ the apparent viscosity decreases with increasing shear rate and the fluid is called pseudoplastic or shear thinning.

2.2 Geometry designs of delivery device and grid generation

When cells are delivered with a syringe device, they pass through the syringe barrel to the needle, which has a much smaller diameter. This sudden contraction causes an increased velocity of the fluid within the needle which could damage the cells. Therefore, the transition from the syringe barrel to the needle is critical for the design of the delivery device, in order to ensure cell viability.

Syringes with different needle diameters and different geometries were designed in SolidWorks®. Four different needle diameters were tested, with the larger diameter being 20 Gauge and the smallest 26 Gauge (Table 1). The straight needles were made to emanate co-axially from a syringe barrel ($D = 1.457$ mm inner diameter), the geometry of which was kept constant in all designs. The angle between the syringe barrel and the attached needle was also kept constant at 45^o.

Three different geometries were designed, based on low dead space syringes (Figure 1). This type of syringes limit the dead space between the syringe hub and the needle, and are thus widely used for medical applications since they ensure accurate dosing. For all three different geometries, the syringe barrel (of inner diameter D) and the diameter of the needle (24 Gauge) were kept constant. The angle in the entrance from the syringe

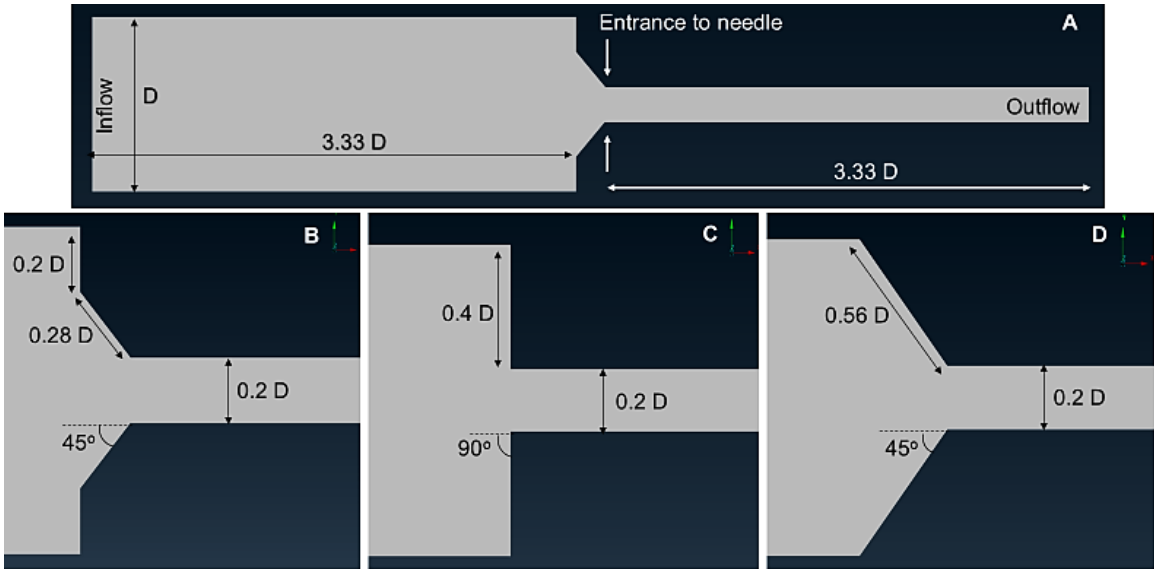

Figure 1: Geometry designs of the low dead space syringe device. A) The syringe consists of the syringe barrel (left) and the needle (right). $D = 1.457mm$ for the inner diameter of the barrel, while the needle diameter was 24 Gauge ($0.2D$), for all geometries. B) Geometry 1: Common design of a low dead space syringe (Angle at the entrance to the needle is 45^o). C) Geometry 2: Angle to the needle entrance is 90^o. D) Geometry 3: Angle to the needle entrance is 45^o.

Table 1: Needle diameter dimensions

Needle Diameter (Gauge)	Needle Inner Diameter (mm)	Needle Inner Diameter (D)
20	0.603	$\sim 0.41D$
22	0.413	$\sim 0.28D$
24	0.311	$\sim 0.21D$
26	0.260	$\sim 0.18D$

barrel to the needle was 45^o in Geometries 1 and 3 (Figures 1B and 1D, respectively), and 90^o in Geometry 2 (Figure 1C).

The ANSA pre-processor (BETA CAE Systems) was used to create a high-quality grid using quadrilateral elements in order to enhance the modelling of the viscous flow. The region where the flow enters the needle is of particular interest, therefore a finer mesh was created in this area. A finer mesh was also created near the walls to capture the near-wall viscous effects (Figure 2A).

In order to achieve reliable simulations, it is important that the results do not depend on the grid. A sensitivity analysis was thus conducted to compare how the velocity varies depending on an increasing number of elements (Figure 2B). The geometry of the 22 Gauge needle was used for that purpose, and the velocity profiles were compared at the entrance of the flow to the needle. It was shown that the solution of the maximum velocity magnitude at the entrance to the needle reached a plateau as the number of elements in the grid increased. The difference between a mesh with 6000 elements in total and a finer

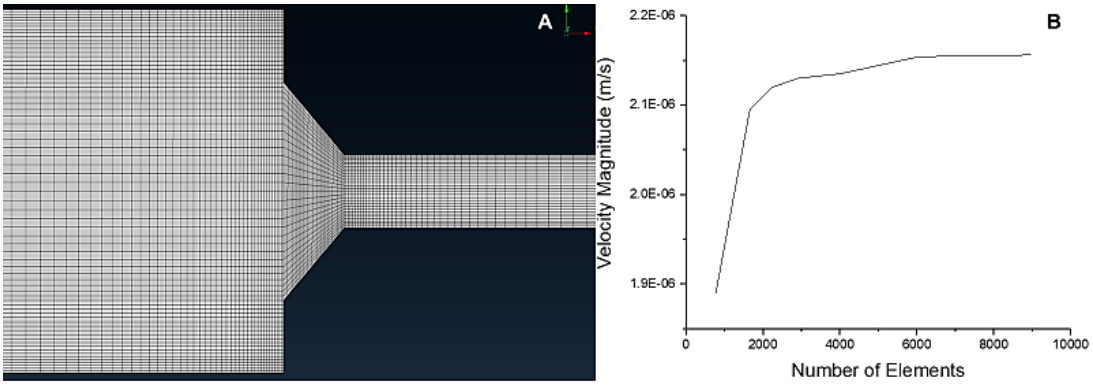

Figure 2: A) Detail of the mesh used for the simulations. Finer mesh was created near the walls and at the entrance to the needle to capture viscous effects. B) Grid dependency analysis based on the maximum velocity magnitude at the entrance of the needle depends on the total number of elements on the grid. The value becomes independent for grids consisting of a total number of elements of 6000 or higher.

mesh with 9000 elements in total was insignificant, thus a mesh of an average of 6000 elements was used in all simulations to reduce the computational cost.

2.3 Computational modelling of collagen flow in syringes

Due to the low concentration of collagen, the flow was considered incompressible and was simulated using a finite volume approach in OpenFOAM®, utilising the SIMPLE algorithm. The mass conservation law for an incompressible fluid is given by the equation:

$$\nabla \cdot \vec{u} = 0 \tag{3}$$

The conservation of momentum is given by:

$$\rho \frac{\partial \vec{u}}{\partial t} + \rho(\vec{u} \cdot \nabla)\vec{u} = -\nabla \mathrm{p} + \mu \nabla^2 \vec{u} \tag{4}$$

where $\vec{u} = [u, v, w]$ is the velocity vector, ρ is the fluid density, p is the static pressure and μ is the dynamic viscosity.

The non-Newtonian fluid constitutive behaviour that was characterised experimentally was modelled using the power law model. A steady state volumetric flow rate of 4 $\mu l/min$ was used in the simulations, with a constant inlet velocity corresponding to maximum delivery volume. The inlet velocity profile was assumed uniform which is a reasonable assumption for the case of a syringe plunger and a highly viscous collagen solution.

3 RESULTS

3.1 Collagen shear thinning behaviour

In non-Newtonian fluids, viscosity depends on the magnitude and the rate of the applied shear stress. For materials with shear thinning properties, the viscosity decreases as the shear rate increases. In this study, an investigation to confirm whether soluble collagen in low concentrations ($2mg/ml$) exhibits shear thinning properties was conducted. The apparent viscosity (in cP) was examined against the shear rate (in s^{-1}) and the results

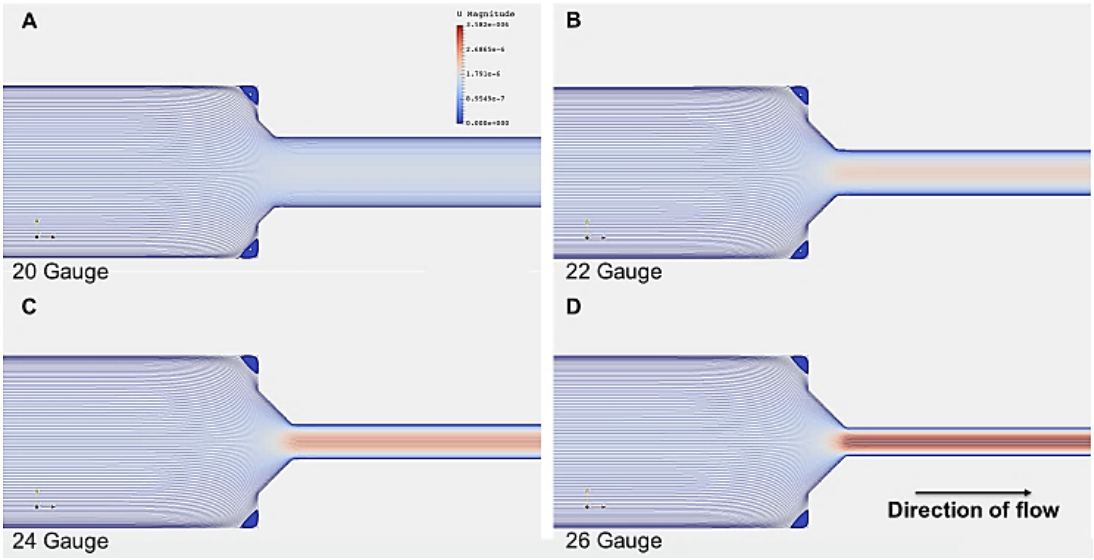

Figure 3: Velocity streamlines on syringes with different needle diameters. Low dead space syringe with a (A) 20, (B) 22, (C) 24, and (D) 26 Gauge needle attached. (A) is currently used in intrastriatal injections, while (C) is almost half in diameter of (A) and could reduce the risk of hemorrhage, but possibly damage the cells; (D) is smallest needle diameter. Streamlines coloured by velocity magnitude based on the colourmap in (A). Flow is from left to right.

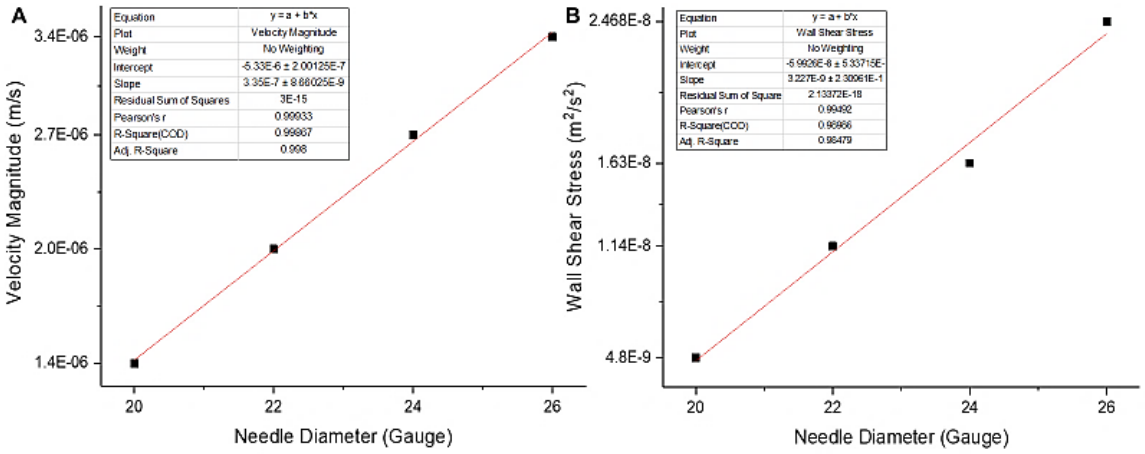

Figure 4: Almost linear relationships between the needle diameter and (A) the maximum velocity magnitude and (B) the wall shear stress at the entrance to the needle.

were analysed using the power law model. For low concentrated soluble collagen, the flow behaviour index, n, was lower than 1 ($n = 0.903$). Even though this indicates a shear thinning fluid, the material's behaviour was not far from being characterised as Newtonian.

3.2 Comparison of Newtonian and non-Newtonian model

The flow of collagen in a syringe with 24 Gauge needle diameter was simulated in OpenFOAM®, as described in section 2.3, for Geometry 1 (Figure 1B). Results were

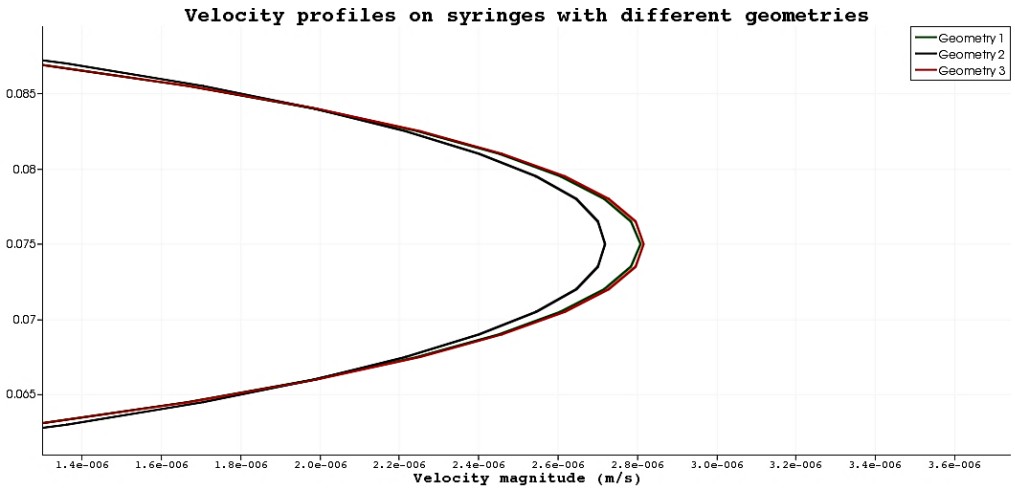

Figure 5: Velocity profiles at the entrance of the needle, indicating differences in the pressure drop in the needle fitting for Geometry 2 as compared to Geometries 1 and 3, for the same inlet flow rate.

acquired for a shear thinning fluid, described by the power law, and a Newtonian fluid model. It was found that the maximum velocity at the entrance of the needle was almost identical for both the Newtonian and the non-Newtonian fluid models. No statistical difference was found in the velocity profiles, indicating that collagen's behaviour in low concentrations is very similar to a Newtonian fluid. Therefore, all remaining simulations were performed with the assumption of a Newtonian fluid.

3.3 Effect of needle diameter on collagen flow

One of the main concerns in intrastriatal injections is the possible tissue damage and hemorrhage that may occur. As a standard practice, needles with 18 to 20 Gauge diameter are used to deliver therapeutics to the brain. However, as this may cause damage to the site of the delivery, it would be beneficial to use needles with a smaller diameter.

Four different needle diameters were tested, of 20, 22, 24, and 26 Gauge (Figure 3) to establish the effect of needle diameter on the flow of the collagen. Not surprisingly, an almost linear relationship was observed between the needle diameter and the maximum velocity magnitude at the entrance to the needle (Figure 4A). The shear stress on the wall was also analysed. Highest wall shear stresses were observed at the needle entrance, exhibiting an almost linear relationship with increasing needle diameter (Figure 4B).

3.4 Effect of syringe geometry on collagen flow

In many medical applications, low dead space syringes are considered a quality standard since they limit the dead space between the syringe hub and the needle, thus ensuring accurate dosing. However, the design of a low dead space syringe can vary. In this study, the effect of three different geometry designs on the flow of collagen was investigated. It was observed that the maximum velocity magnitude at the entrance of the needle was similar for Geometries 1 and 3, but lower for Geometry 2 (Figure 5), for the same inlet flow rate. This indicates differences in the pressure drop in the needle fitting for each

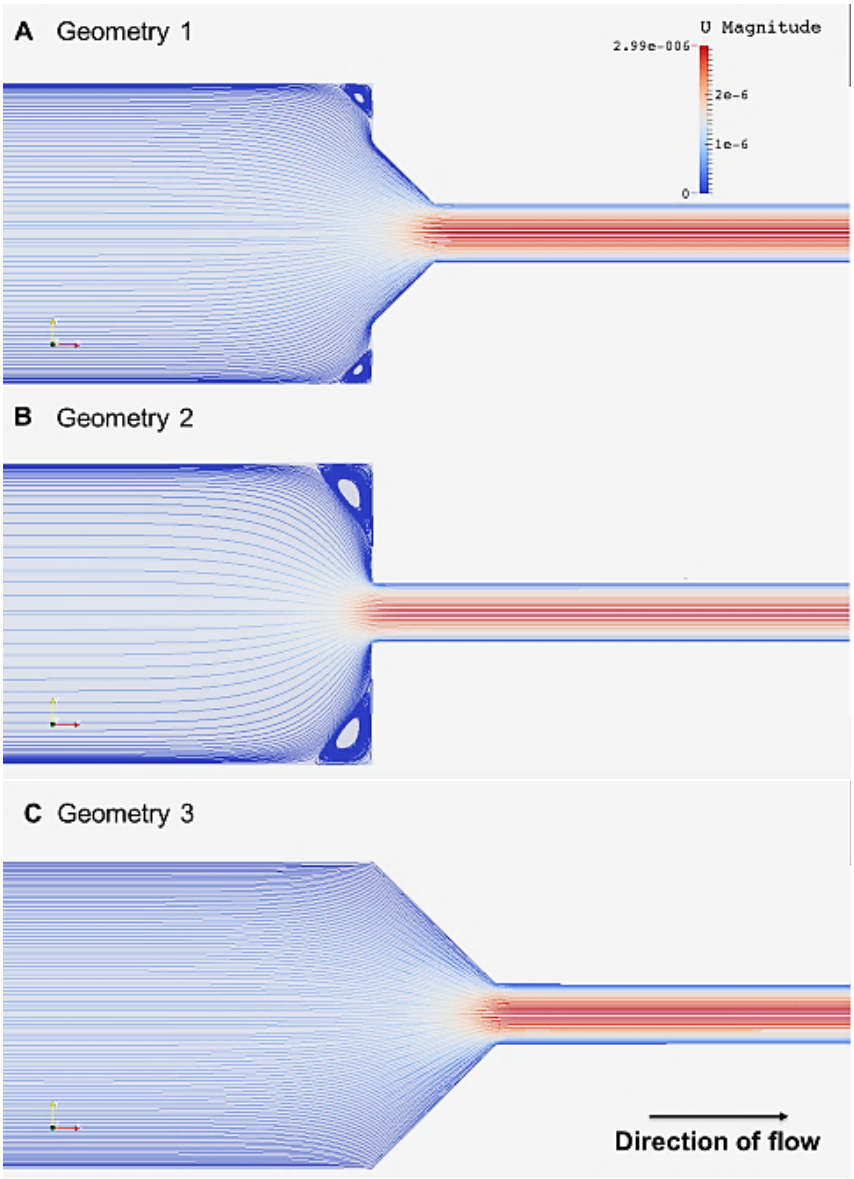

Figure 6: Velocity streamlines for different geometry designs of low dead space syringes, using a 24 Gauge needle. A) Geometry 1: Recirculation zones appear at the corners of the syringe barrel. B) Geometry 2: Stronger recirculation zones appear due to the more sudden contraction. C) Geometry 3: No recirculation zones present.

syringe Geometry.

The lower maximum velocity magnitude at the needle entrance for Geometry 2 could be attributed to the higher vorticity being observed at the corners of the syringe barrel (Figure 6). Recirculation zones appeared at the corner ends of syringe Geometry 1 and Geometry 2, with the strongest being for Geometry 2. These vortices could prove a disadvantage on these syringe designs since they could possibly cause cell entrapment during the delivery. No recirculation zones were apparent for Geometry 3.

Finally, the shear stress on the wall of the syringes was measured. The maximum

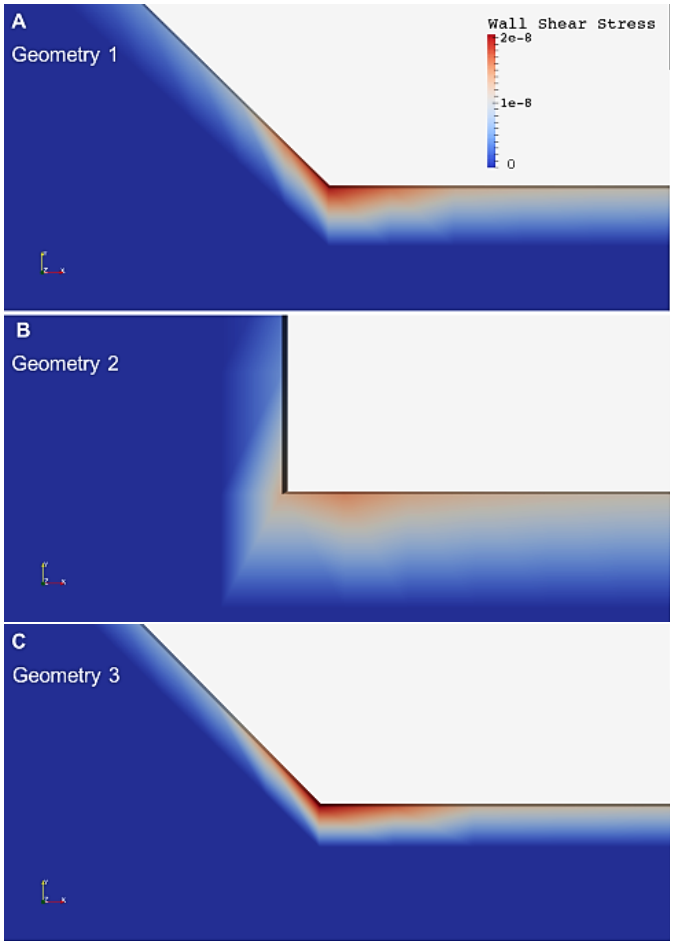

Figure 7: Contours of wall shear stress (WSS) near the entrance to the needle with maximum values being observed at the corner where the fluid entered the needle. A) Geometry 1. B) Geometry 2 had the lowest WSS values compared to the other two designs (35% lower than Geometry 1 and 40% lower than Geometry 3). C) Geometry 3. Flow is from left to right.

Table 2: Maximum wall shear stress on syringes with different designs

Geometry	Maximum Wall Shear Stress (m^2/s^2)
1	1.95E-08
2	1.25E-08
3	2.05E-08

shear stress was measured at the corner region where the needle was attached to the syringe barrel (Figure 7). The overall maximum wall shear stress value was 35% lower on Geometry 2 compared to Geometry 1 and 40% lower compared to Geometry 3 (Table 2). However, since the same needle diameter was used for all three designs, this difference on wall shear stress was observed at the entrance of the flow to the needle; once the flow became fully developed further downstream of the needle entrance, no differences on the wall shear stress were noticed.

4 DISCUSSION AND CONCLUSIONS

This study highlights the effect of geometry and needle diameter on the design of new medical devices for cell delivery to the CNS. As collagen passes from the syringe barrel to the needle, variations in the velocity, pressure drop, and wall shear stress are observed due to different needle diameters and different barrel geometric designs.

Collagen, a material widely used in biomedical applications, has been characterised with non-Newtonian behaviour [15]. In previous studies, its shear thinning behaviour has been identified, which is a desirable characteristic for applications as an injectable scaffold since it can facilitate the injection to the site [16]. However, in this study, low concentrations of soluble collagen were studied, and when compared to higher concentration of soluble collagen and collagen gels, the material behaviour was similar to Newtonian fluid. The implementation of the power law model in the computational analysis did not affect the results, which were comparable to the Newtonian model. Collagen, however, is a complex material. Collagen structure consists of three polypeptide chains, coiled together to form a triple helix. Three of these helices then form a molecule of tropocollagen, the basic building block of collagen. Finally, tropocollagen molecules aggregate in a staggered fashion to form collagen fibrils which are stabilised mainly by covalent cross-links [17, 18]. Therefore, further analysis is required to identify how these structures behave under shear stress, specifically during injection.

When cells flow through a syringe needle, they experience fluid forces that can lead to cell disruption and even cell death [19]. These forces include pressure drop across the cell, shearing forces due to shear flow and stretching forces due to extensional flow [20]. In previous studies, it was shown that extensional flow is the major cause of cell death during an injection [13]. This is more likely to occur when higher Gauge needles are used, since the stretching forces would be higher due to the augmented velocity observed in smaller diameters. Current approaches utilise needles with 18 to 20 Gauge diameter for the delivery of therapeutics to the CNS. The cell viability during injection has been investigated in needles with a bigger diameter, and no correlation between cell viability and needle diameter was observed [12]. However, this is within certain limits, since needles with a maximum of only 22 Gauge were tested. Therefore, it would be beneficial to identify the limit above which cell viability would be compromised. This could help ensure the successful delivery of a cell therapy to the CNS with a smaller needle, which would be preferred since it can minimise the tissue damage at the site of the delivery and can reduce the haemorrhage risks.

Finally, of main consideration is the design of the syringe barrel. Current approaches utilise low dead space syringes, which are nowadays considered a medical standard, since they allow accurate dosing. However, different geometric designs can be used to achieve the low dead space between the needle and the syringe hub, and it would therefore be of interest to investigate how these designs can affect the fluid flow. In a previous study, it was shown that needle geometry can affect the cell delivery, with a tapered needle minimising the cell damage compared to a cylindrical needle [21]. This observation was attributed to the change in force distribution between the two geometric designs. In the

present study, the effect of the syringe barrel design to the fluid flow was investigated and a geometric design with a more sudden contraction between the syringe barrel and the needle was shown to reduce the maximum velocity magnitude at the entrance to the needle, indicating different pressure drop along the syringe-needle connection. The wall shear stress was also shown to be lower at the same site of the syringe, which could be attributed to the stronger vorticity that was observed in this design.

Further analysis is required including the simulation of cells during injection and analysis of their deformation, in order to ensure that the device is designed to provide the optimum viability to the cells delivered. The design and development of a medical device that would ensure the viability of the cells delivered, while minimising the tissue damage and the immune response observed during intrastiatal injection could help bring the stem cell research for neurodegenerative diseases to a clinical translation.

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