Alginate flow seeding microparticles for use in Particle Image Velocimetry (PIV)

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ABSTRACT

Appropriate flow seeding is particularly critical with in the particle image velocimetry (PIV) technique that indirectly measures the displacement from the particle velocity instead of fluid velocity. Particles that follow the flow accordingly and scatter enough light must be used in order to obtain accurate velocity field of the flow. Therefore, particles should be as small as possible in order to ensure good tracking of the fluid motion and they should not be too small, since they will not scatter enough light. The microparticles can be obtained from approximately 20 nm to around 50 μm in a controlled manner with ability increasing their light scattering behavior by fluorescein imbedding so that they gain the light scattering property.
Alginate microparticles as flow seeding fulfill all the requirements that are recommended for the velocity measurements in PIV. These spherical microparticles offer the advantage of being environmentally friendly, excellent seeding properties and very simple production ability. They can be obtained as small as possible while increasing its scattering behavior by adding fluorescein into them. Additionally, they can be perfectly used as flow seeding in gas and several liquid flows with matching densities.

INTRODUCTION

Micro and nanoparticles are used in wide range of flow analysis of the particle image velocimetry (PIV) applications like single-phase liquid flows, industrial large-scale flows, near-wall flows, turbulent flows, in turbo-machinery flow and complex flow systems [1]. Descriptions and characteristics of seeding particles can be found in the literature [2]. Selection process of the particles comprises a basis for the velocity measurements in PIV. These particles should be as small as possible in order to ensure good tracking of the fluid motion. On the other hand, they may not be too small, since they will not scatter enough light. Particles that follow the flow accordingly and scatter enough light must be used in order to obtain accurate velocity field of the flow.

When applying the PIV technique to measure the flow velocity, the area interest containing the fluid and the seeding particles is illuminated at least twice within a short interval by means of a laser in a plane of the flow. It is assumed that tracer particles move with the local fluid velocity between the two illuminations. The light scattered by the particles is recorded on a single frame or on a sequence of frames. The displacement of the particle images between the light pulses is then determined through evaluation of the PIV recordings. The technique indirectly measures the displacement from the particle velocity which is determined instead of fluid velocity. Therefore, fluid mechanical properties of the particles have to be checked in order to avoid significant inconsistencies between fluid and particle motion [3].

The most commonly used seeding particles for PIV investigations of liquid flows are polystyrene, aluminum, glass spheres as solid seeding, different types of oils as fluid seeding and oxygen bubbles as gaseous seeding, [2, 3]. Resulting particle image intensity is directly proportional to the scattered light power. Therefore, it is more effective and economical to increase the image intensity by properly choosing the scattering particles than by increasing the power of the laser. The scattered light by small particles is a function of the ration of the refractive index of the particles to that of the surrounding medium, the particle's size, their shape and orientation. There is a tendency for the scattered light intensity to increase with increasing particle diameter. However, increasing the size of the particles will affect tracking of the fluid motion. Moreover, difficulties arise in providing high quality seeding in gas flows compared to applications in liquid flows. The most commonly used seeding particles for PIV investigations of gaseous flows are polystyrene, aluminum, magnesium, glass micro-balloons as solid seeding and different oils as liquid seeding [2]. Therefore, it is clear that a compromise to use seeding particles has to be found for liquid and gaseous flows.

Alginate is the most popular material that is used for preparation of gel beads among the wide range materials [4, 5]. It is a natural, non-toxic polysaccharide found in all species of brown algae [6]. Chemically alginate is a linear 1, 4 linked copolymer of β-D-mannuronic acid (M) and α-L-guluronic acid (G). Alginates are widely used in the food and pharmaceutical industries and have been employed as a matrix for the entrapment of drugs [7], macromolecules [8, 9] and biological cells [10, 11].

Alginate microspheres are generally prepared by two alternative methods; (i) by dropping aqueous alginate into a solution of calcium salt [12, 13] or (ii) by emulsification method under gentle stirring [14, 15]. The particles that are produced by (i) are generally larger (more than 1 mm in diameter) and the attempts to obtain smaller particles requires special device that can have the disadvantage of the high cost and possible clogging [16]. The (ii) method has recently been extended to the field of nanotechnology. Experimental evidence shows that alginate nanoparticles have been successfully prepared using this method by creating the ideal conditions for the formulation [17]. Some formulation and process parameters have been adjusted to produce smaller particles. The emulsification/internal gelation procedure provides an appropriate way to produce the alginate microspheres with a preferable diameter sizes. Entrapment within alginate spheres is considered as safe and simple system with good mechanical stability [18]. By controlling the experimental conditions, size can be easily controlled from a few nanometers to millimeters in diameter [19]. In internal gelation, calcium ions are homogeneously distributed in the alginate solution, thus diffusion of protons into the pre-gel droplets induce gelation starting at the surface, giving rise to homogenous droplets [20]. Also, the low shear involved in internal gelation protects fragile encapsulants [21]. In addition, since toxic reagents and solvents are not used, biological and food applications may also be considered. This method permits the efficient production of large quantities of small and controlled diameter alginate micro- and nanoparticles. The alginate microparticles that are produced by this method are more porous than externally gelled particles. This situation is usually considered a disadvantage depending on the application. It can serve as an advantage with the PIV system due to the nature of PIV requiring less difference in matching fluid-particle densities.

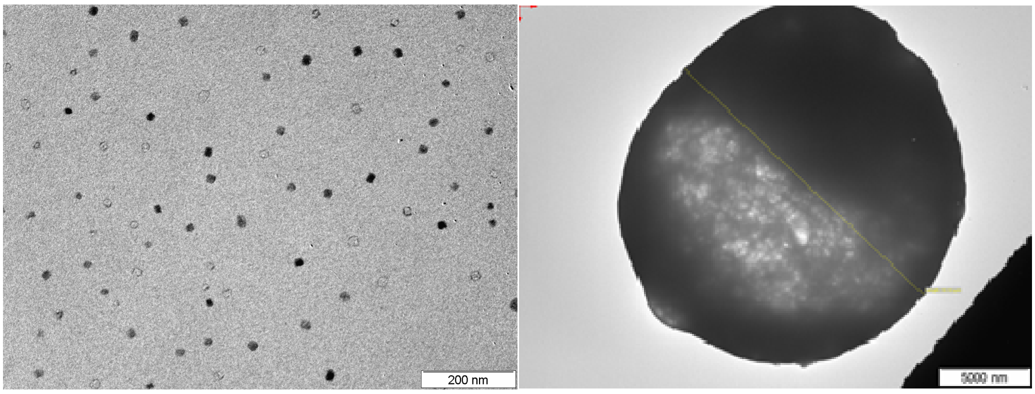
In the present study, alginate micro and nanoparticles have been developed and characterized as a new alternative of conventional seeding particles for the velocity measurements in PIV. Two PIV applications, water through a gear pump system and air through a computer fan, have been conducted to check the performance characteristics of the particles. Developed particles offer the advantages of being environmentally friendly, excellent flow seeding and light scattering abilities. They ensure good tracking of the fluid motion in both liquid and gas flow measurements. When they are used in liquid, there is not too much difference in the density between the fluid and the particles thanks to their porous structure that easily absorb the analyzing fluid. Moreover, the microparticles can be obtained from approximately 20 nm to around 50 μm in a controlled manner with ability increasing their scattering behavior by imbedding fluorescein into them so that they gain the light scattering property.

Alginate gel micro and nanoparticles are prepared by the emulsification/internal gelation method adapted from previous reports [22, 23, 24, 25] for encapsulation of the particles. In order to obtain smaller particles, typically $d < 10$ μm, a tip ultrasonicator is used. This is possible due to the formation of the water-in-oil type emulsion, where droplets are stabilized by the use of surfactants. For the experiments that are conducted with green Nd-Yag laser, the particles are prepared by adding fluorescein into aqueous solution. In this way, alginate particles that encapsulated fluorescein are formed with the same procedure as mentioned above.

RESULTS AND DISCUSSION

Characterization

To characterize the alginate particles the transmission electron microscopy (TEM), environmental scanning electron microscopy (ESEM) and confocal laser electron microscopy techniques have been used. The diameter size of the microparticles is increased when they are in the analyzing fluid due to the porous property of the particles. Figure 1a) shows TEM results of the dry and partially dry nanoparticles. Some of the microparticles have appeared completely black and the rest of them had black circles with white zone inside. The black ones refer to the microparticles that are partially wet. The diameter size of the dried microparticles has been obtained approximately 20 nm. This size is changing when the microparticles are in liquid. Because of their porous structure, they absorb the liquid. This leads to increase the diameter size of the microparticles from 5 μm up to 35 μm depending on the absorbed liquid. Figure 1b) shows TEM results of alginate microparticles that have absorbed the liquid. The diameter size of the microparticles has been reached to 19.18 μm. TEM results are not definite results in order to verify the explanation of microparticles how are changing in diameter size. From the TEM images, we can state that this is only a confirmation when the microparticles are in liquid; they absorb large amount of liquid with an extensive area.



a) **b)**

Figure 1 **a)** TEM results of dry alginate nanoparticles in 200 nm scale. **b)** TEM results of wet alginate microparticles in 5000 nm scale

From the Figure 1b), it can be observed that alginate microparticles consist of porous structure. This leads to microparticles have the jelly-like structure. This also leads to microparticles have the density of liquid that is the analyzing liquid. This property is significantly important in the PIV technique. The desired seeding particles for PIV should have almost the same density or very similar density with the analyzing liquid. In this point of view, alginate microparticles seem to be one of the most adequate tracer particles for PIV.

Figure 2 shows TEM images that have been captured in small section areas. When they are dried, they seem in a very few amount. They have a diameter size of approximately 85 nm. It is a dry case situation. When the microparticles are in water, they absorb and retain water because of their hydrophilic property. The moment that they start to absorb water, images of the microparticles are getting darker as shown in Figure 3. Then they start to float while the rest of microparticles do not show any movement. The diameter size increases approximately from 20 nm to 20 µm (see Figure 1b)). This condition leads to obtain bigger particle sizes in order to illuminate the microparticles by laser and leads them to be seen in the flow with an almost exact density of the water in order to obtain a good tracking. This situation is an excellent advantage for PIV systems.

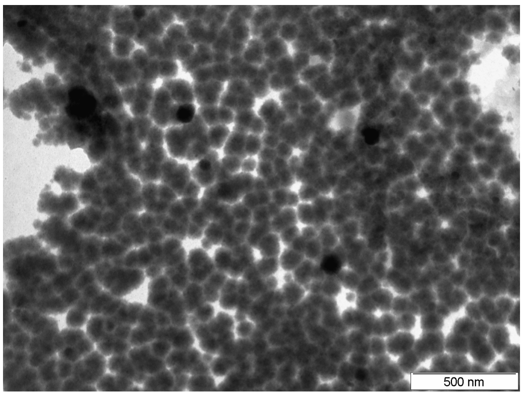
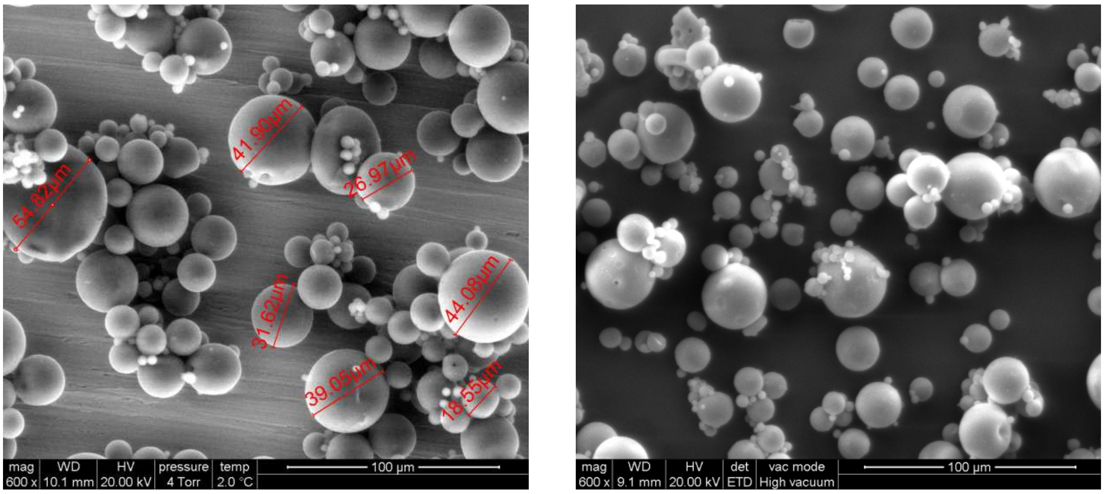


Figure 2 TEM results of dry and partially dry (black dots) alginate microparticles in a 500 nm scale.

ESEM images of the microparticles are shown in Figure 3 in order to compare the in liquid and dry form. They have been filtrated in dry form by using a 50 µm sieve. The microparticles have been obtained with a bigger diameter sizes in liquid form because of their hydrophilic property. Furthermore, the microparticles have an almost perfect shape as spheres in liquid and dry form.



a) **b)**

Figure 3 ESEM images of alginate microparticles in **a)** liquid and **b)** dry form in 100 µm scale.

Confocal laser electron microscopy has been used to investigate the microparticles that contain fluorescein. Figure 4 illustrates a 3D image that has been captured with blue laser which has a 488 nm wavelength. It can be observed that the average particle diameter is less than 30 µm. This diameter size can be efficiently used for tracer microparticles in PIV experiments. The fluorescein that is appended to the alginate microparticles leads to the PIV system's tracer microparticles a highly light scattering property. Again as we can see in Figure 4, a high amount of fluorescein is gathered and attached to the microparticles and particle shape is rounded. PIV technique takes the advantages of these properties of the microparticles and uses efficiently in the flow systems to obtain accurate digital images.

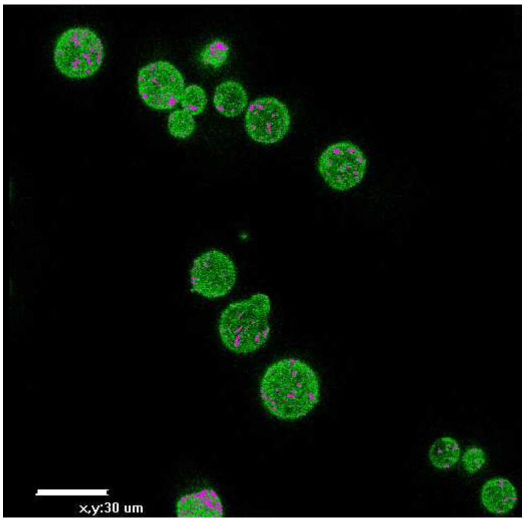


Figure 4 Confocal microscopy images of alginate microparticles that contain fluorescein at 488 nm wavelength.

PIV Experiments

Figure 5 shows that the microparticles have enough light to scatter when a pulsed infrared diode laser with a wavelength of 800 nm has been used as illumination. The alginate microparticles that do not contain fluorescein are very sensitive to the infrared laser. It was not possible to obtain a proper image by using a green laser. However, the microparticles can be adequately used as flow seeding with illumination by an infrared laser.

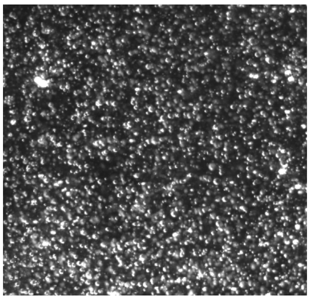
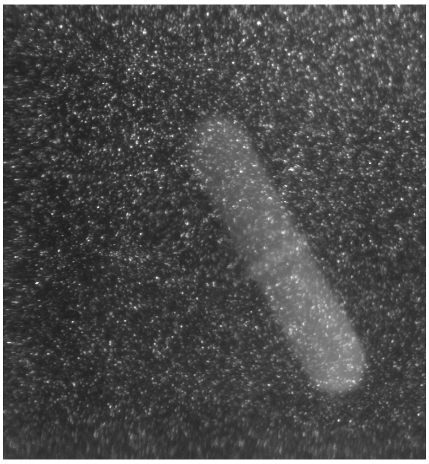
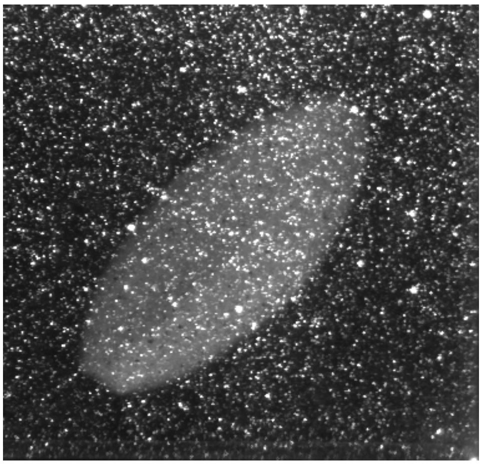


Figure 5 PIV experiment of the alginate microparticles, engine oil with density of 885kg/m^3 and viscosity of 0.028 μm has been used as liquid and pulsed infrared diode laser with a wavelength of 800 nm has been used as illumination.

In order to increase light scatter quality of the microparticles by using green laser, very small amount of fluorescein has been added into the alginate microparticles. The production process remains the same. Fluorescein can be excited at wavelengths as 515 nm. This wavelength range can be given by green Nd-Yag laser. This means that the fluorescein contained alginate microparticles can be used as a better alternative of tracer microparticles for green lasers. Figure 6 shows two examples of PIV images which are recorded at the same conditions of camera and laser. The analyzing fluids are; water for Figure 6a), and olive oil for Figure 6b).



a)



b)

Figure 6 Illumination of alginate microparticles contains fluorescein by using NdYAG green laser with a wavelength of 532 nm **a)** Distilled water has been used as liquid, **b)** Olive oil has been used as liquid

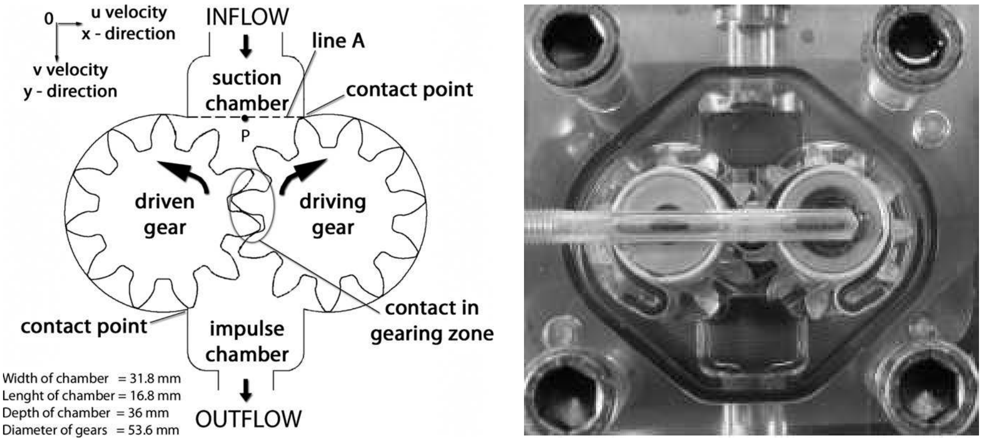
Figure 6 shows homogenous and clear microparticles flow area. Thus, it can be stated that fluorescein contained alginate microparticles can be used with green and infrared diode lasers. It should be noted that only 40-50 % power of green and infrared lasers is used to obtain PIV images that are illustrated in both Figures 7 and 8. In addition, very few amounts of microparticles have been used for these systems. For the system in Fig. 6a) 0.06 g of alginate microparticles contains fluorescein is used with a 600 g of distilled water and in Fig. 6b) 0.04 g of alginate microparticles contains fluorescein is used with a 93 g of olive oil.

Alginate microparticles are porous structures and dispersion of fluorescein outside to alginate microparticles is a feasible situation. To obtain PIV images, this dispersion is not playing an important role whether microparticles lose their scattering light property. Thus, alginate microparticles that contain fluorescein can be used until the time that they are not illuminated by green laser.

The experimental setup of the gear pump system is previously described [26]. The air flow experiments are done in a system consisting of a 9 cm diameter air fan with infra-red laser.

PIV application: Gear Pump

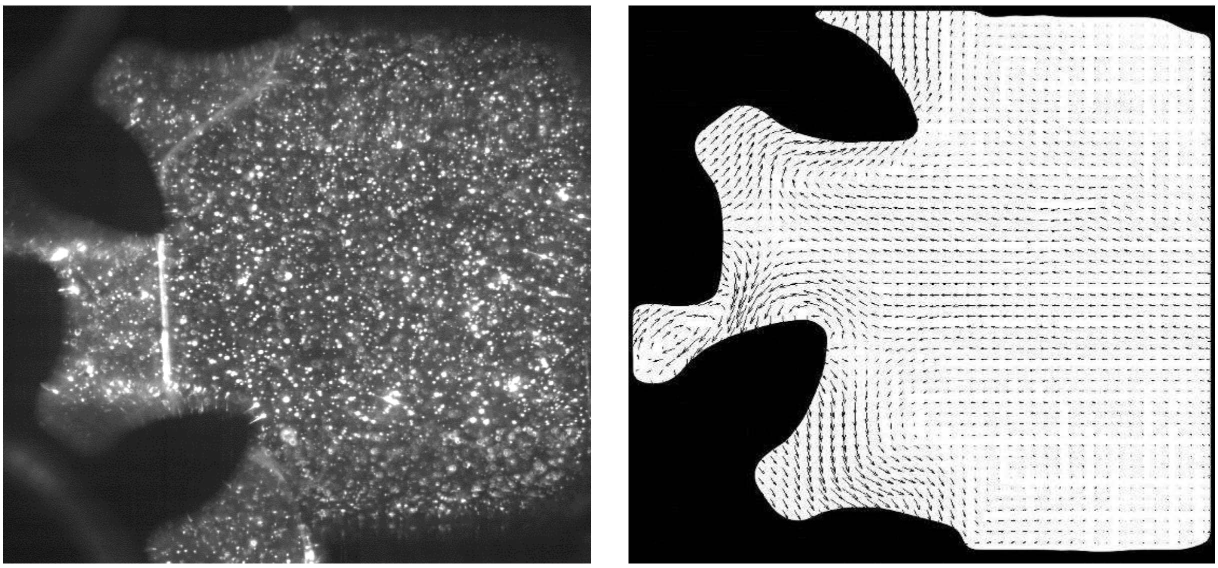
There are special cases of the PIV system that require using appropriate tracer microparticles. As an example; in the analysis of an external gear pump, solid and water droplets were not suitable particles as flow seeding due to possible damages to the external gear test pump [27].



a) **b)**

Figure 7 **a)** Schematic drawings of the gear pump system. Line A and point P indicate the selected location where specific analyses have been done, **b)** external gear test pump, top view.

For this reason, the authors have been used air-bubbles as flow seeding in the PIV experiments. However, controlling the diameter size of the air bubbles was quite difficult. We have previously reported flow analysis of the suction chamber by PIV technique by using air micro bubbles as flow seeding [26]. Particularly this specific external gear pump system led us to seek new tracer particles. Alginate microparticles as flow seeding fulfill the demand of PIV requirements for this gear pump system. A new advantage has been introduced to this project thanks to smaller diameter sizes, approximately 10µm (In the previous report, the diameter of the air bubbles was approximately 100 µm). This advantage leads to obtain a higher spatial resolution of the particle images (see Figure 8).



a) **b)**

Figure 8 **a)** Illumination of alginate microparticles in the suction chamber of an external gear pump. Analyzing fluid is a commercial oil with a density of 885 kg/m^3 and viscosity of 0.028 Pa·s , **b)** Corresponding velocity vectors

PIV application: air flow

The alginate microparticles have been tested in air flow as well. A wind tunnel with velocity of 40 m/s has been used (Figure 9). The microparticles that contain fluorescein have been used as flow seeding and a green laser has been used as light source. Figure 10 illustrates an example of PIV image that has been recorded for air flow.

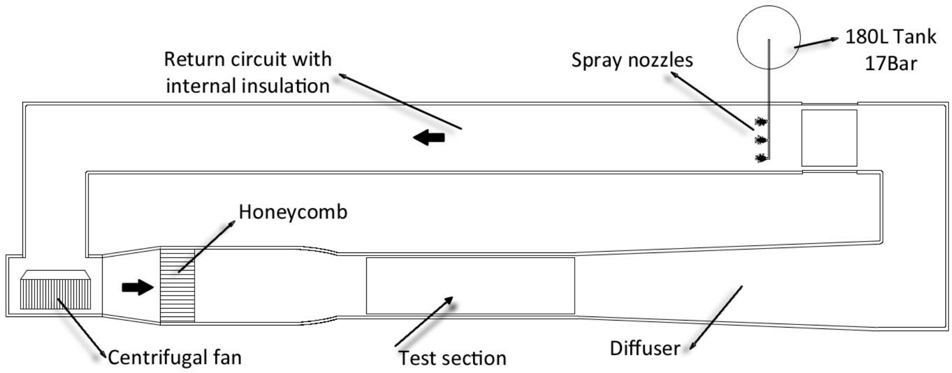


Figure 9 Schematic view of wind tunnel

It has been observed that alginate microparticles that contain fluorescein are very sensitive to the air flow as well. Straight direction of the air flow resulted in very orderly distributed velocity vectors as can be seen in Figure 9b). PIV images have been obtained with smaller size of microparticles which means that spatial resolution of the images can be increased by increasing the amount of microparticles per interrogation area.

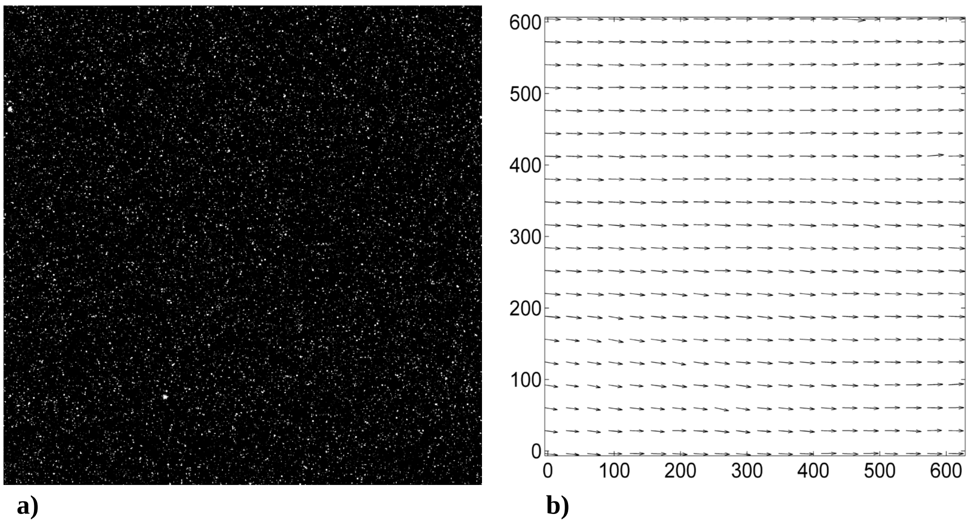


Figure 10 **a)** PIV image of fluorescein contained alginate microparticles (600 x 600 pixels; 30 mm x 30 mm) in air by using Nd-YAG green laser with a wavelength of 532 nm **b)** corresponding velocity vectors.

CONCLUSIONS

The aim of the present study is to produce alginate microspheres with a simple characterization and use them as tracer particles in different fluids. In addition, the study aims to attempt a trend of its use as tracer particles in PIV experiments by improving its properties.

In some biological and food application areas, alginate microspheres that are produced by emulsification method have a disadvantage as being porous structure. This study transforms this disadvantage to an advantage. In the use of PIV experiments, porous structure leads to obtain density which is almost the same with analyzing fluid. This means that the microparticles follow the analyzing fluid efficiently.

Fluorescein has been added into the alginate microparticles that gives them a better property of light scattering by using green laser at the same time. It is certain that microparticles will carry efficiently the property during the PIV experiment. Furthermore, alginate microparticles with fluorescein can be used as flow seeding in PIV technique for liquid and gas flows analysis.

Alginate microparticles have soft, jelly like structure. Thus they do not cause any damage to the inner contact surface of the machines. So that the microparticles are very appropriate for turbo-machinery applications in PIV.

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REFERENCES

[1] Choi HM, Kurihara T, Monji H, Matsui G, "Measurement of particle/bubble motion and turbulence around it by hybrid PIV" Flow Measurement and Instrumentation 12 (2002) pp.421-428
[2] Adrian RJ "Twenty years of particle image velocimetry" Experiments in Fluids 39 (2005) pp.159-169
[3] Raffel M, Willert CE, Kompenhans J, "Particle image velocimetry: a practical guide" Springer Berlin (1998)
[4] Mattiason B "Immobilized cells and organelles" CRC Press Boca Raton FL (1983)
[5] Veliky IA and Mclean RJC "Immobilized biosystems theory and practice applications" Backie and Professional, London (1994)
[6] Aslani P and Kennedy RA "Studies on diffusion in alginate gels. I. Effect of cross-linking with calcium or zinc ion on diffusion of acetaminophen" Journal of Controlled Release 42 (1996) pp.75-82
[7] Tomida H, Mizuo C, Nakamura C, Kiryu S, "Imipramine resalease from Ca-Alginate gel beads" Chemical and Pharmaceutical Bulletin 41 (1993)pp.1475-1477
[8] Kim CK and Lee EJ "The controlled release of blue dextran from alginate beads" International Journal of Pharmaceutics 79 (1992) pp.11-19
[9] Mumper RJ, Huffman AS, Puolakkainen PA, Bouchard LS, Gombotz WR., "Calcium-alginate beads for the oral delivery of transforming growth factor-β1 (TGF-β1): stabilization of TGF-β1 by the addition of polyacrylic acid within acid-treated beads" Journal of Controlled Release 30 (1994) pp.241-251
[10] Lim F and Sun A "Microencapsulated islets as bioartificial endocrine pancreas" Science 210 (1980) pp.908-910
[11] Lee GM and Palsson BO "Stability of Antibody Productivity Is Improved When Hybridoma Cells Are Entrapped in Calcium Alginate Beads" Biotechnology and Bioengineering 42 (1993)pp.1131-1135
[12] Salib NN, El-Menshawy MA, Ismail AA, "Utilization of Sodium Alginate in Drug Microencapsulation" Pharmazeutische Industrie 40-11A (1978) pp.1230-1234
[13] Matsumoto S, Kobayashi H, Takashima Y "Production of monodispersed capsules" Journal of Microencapsulation 3 (1986) pp.25-31
[14] Wan LSC, Heng PWS, Chan LW "Development of alginate microcapsules by emulsification" Proceedings of the NUS-JSPS Seminar on Recent Developments in Pharmaceutics and Pharmaceutical Technology (1990) pp. 243-255.
[15] Poncelet D, Poncelet De Smet B, Beaulieu C, Huguet ML, Fournier A, Neufeld RJ "Production of alginate beads by emulsification/internal gelation. II. Physicochemistry" Applied Microbiology and Biotechnology 43(1995) pp.644-650
[16] Skjâk-Braek G and Martinsen A "Seaweed Resources in Europe: Uses and Potential" Guiry, G. Blunden (Eds.) Wiley Chichester (1991) pp. 219-257.
[17] Reis CP, Ribeiro AJ, Neufeld RJ, Veiga F, "Insulin-loaded alginate nanoparticles obtained by emulsification/internal gelation" XII International workshop on bioencapsulation, Vitoria, Spain: Servicio editorial Universidad del Pais Vasco (2004)pp. 251.
[18] Esquisabel A, Hernandez RM, Igartua M, Gascon AR, Calvo B, Pedraz JL "Effect of lecithins on BCG-alginate-PLL microcapsule particle size and stability upon storage" Journal of Microencapsulation 17 (2002) pp.363-372
[19] Poncelet D "Production of Alginate Beads by Emulsification/Internal Gelation"Annals of the New York Academy of Sciences 944 (2001) pp.74-82
[20] Quong D, Neufeld RJ, Skjåk-Bræk G, Poncelet D "External Versus Internal Source of Calcium During the Gelation of Alginate Beads for DNA Encapsulation" Biotechnology and Bioengineering 57 (1998) pp.438-446
[21] Poncelet D, Lencki R, Beaulieu C, Halle JP, Neufeld RJ, Fournier A "Production of alginate beads by emulsification/internal gelation. I. Methodology" Applied Microbiology and Biotechnology 38 (1992) pp.39-45
[22] Wan LSC, Heng PWS, Chan LW "Drug encapsulation in alginate microspheres by emulsification" Journal of Microencapsulation 9 (1992)pp.309-316
[23] Wan LSC, Heng PWS, Chan LW "Influence of hydrophile-lipophile balance on alginate microspheres" International Journal of Pharmaceutics 95(1993) pp.77-83
[24] Wan LSC, Heng PWS, Chan LW "Surfactant effects on alginate microspheres" International Journal of Pharmaceutics 103 (1994)pp. 267-275
[25] Heng PWS, Chan LW, Wong T. "Formation of alginate microspheres produced using emulsification technique" Journal of Microencapsulation 20 (2003) pp.401-413
[26] Ertürk N, Vernet A, Castilla R, Gamez-Montero PJ, Ferre JA "Experimental analysis of the flow dynamics in the suction chamber of an external gear pump" International Journal of Mechanical Sciences 53 (2011) pp.135-144
[27] Castilla R, Wojciechowski J, Gamez-Montero PJ, Vernet A, Codina E "Analysis of the turbulence in the suction chamber of an external gear pump using Time Resolved Particle Image Velocimetry" Flow Measurement and Instrumentation 19 (2008) pp.377-384