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# Animal Models for Hydrodynamic Gene Delivery

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## 1. Introduction

Hydrodynamic gene delivery (HGD) is an established method of the last decade where macromolecules, non-normally permeable to cell membrane, are delivered intracellular. The basic principle is that a large volume of solution, containing genes or oligonucleotides, RNA, proteins or other compounds, is infused rapidly in circulation to permit the entrance of these substances to parenchymal cells. Excellent review papers are published summarizing the principles, the applications and the progress that has been made in this field<sup>1-10</sup> The aim of this chapter is to describe the basic principles of the hydrodynamic gene delivery, the surgical procedures in all animal models and the reflection of our scope for the future development.

## 2. Basics of hydrodynamic gene delivery

Hydrodynamic gene delivery is developed based in our knowledge on vasculature, fluid properties and cell membrane permeability. Based on the studies of Zhang et al.<sup>1</sup>, Kobayashi et al.<sup>2</sup>, Lecocq et al.<sup>3</sup> and Al-Dosari et al.<sup>5</sup>, the rapid infusion of large solution containing macromolecules, non-normally permeable to cell membrane, generates high hydrodynamic pressure in the circulation refluxing to the target organ. The enlarged perivenous area, by the extravasation of the infused solution, generates high pressure in the exterior of the cells. When this pressure reaches a certain level, defects (pores) are been created on the cell membrane leading to the insertion of the macromolecules intracellular. After a few seconds, while the pressure declines at post-injection period, the defects are restoring, trapping inside the cytoplasm the infused molecules. Finally, the body adapts the volume overload within time and the homeostasis is again balanced. We believe that the sequence of these events occur in all surgical protocols, from tail vein injection in mice to specific organ infusion in large animals.

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The vehicle for the molecules is Normal Saline, Ringer's Solution or Phosphate Buffered Saline and the dosage range from 0.1 to 10 mg/kg, depending on the application. The molecules can be DNA, as plasmid, fragment or artificial chromosome, RNA, single or double stranded, genomic or not, polymers, proteins and small compounds. The main application of the hydrodynamic delivery is the therapy studies, especially genes encoding secretory proteins which can be even isolated and purified. A different area of interest is the gene function analysis in a whole animal instead of a cell culture; the transfection is applied with the hydrodynamic technique and the conclusions are withdrawn from the whole animal. The study of DNA sequences can be achieved with HGD, where promoters, introns, enhancers, suppressors can be delivered intracellular in whole animal. Moreover new animal models are possible to be created for study human diseases, mainly viral due to the fact that they are species specific<sup>5,8,10</sup>.

### 3. Animal models

#### 3.1 Mice

The majority of the research protocols of the hydrodynamic delivery in mice are based on tail vein injection, due to the ease of application and the positive results<sup>11,14-53</sup>. In the mouse tail there are four blood vessels, the lateral ones being the veins. The animal is placed in a restrain box or in light sedation and prior to injection the tail vein should be dilated with a tourniquet, immersed in hot water (40 °C for 1-2 min) (photo 1) or placed in incubator at 37°C for 10-15 min. A 27g-30g needle at a very shallow angle is commonly used, while the injection of small amount of fluid is mandatory for the verification of the correct placement of the needle (photo 2). Then, the solution of a total 1.5-2 ml, equivalent to 8-10% of body weight, is rapidly infused, within 5 to 10 seconds<sup>4</sup>.

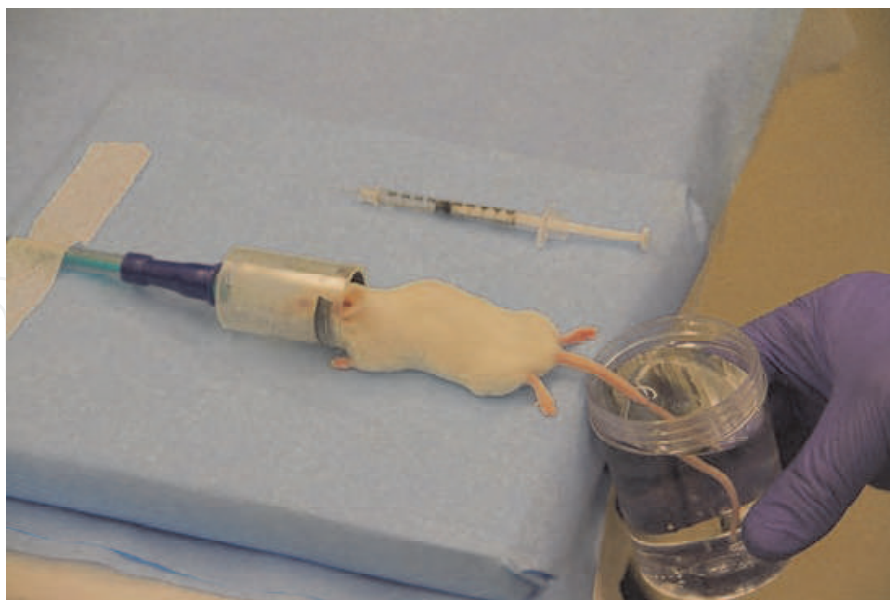


Photo 1. The animal is light sedation with the tail is placed in hot water for dilatation of the veins

A surgical procedure has been established by Zhang et al<sup>15</sup> for delivering macromolecules in liver under general anesthesia. After a midline incision at the abdomen, the portal vein

was identified and the solution was injected with a 30g needle while the hepatic vein and the inferior vena cava had been occluded. Modification of this procedure, by the same group, involved the solution being infused in the hepatic vein with occlusion of the portal vein, vena cava and the hepatic artery. At the end of the infusion gentle pressure was applied at the insertion point to minimize hemorrhage. The midline incision was closed in standard technique.

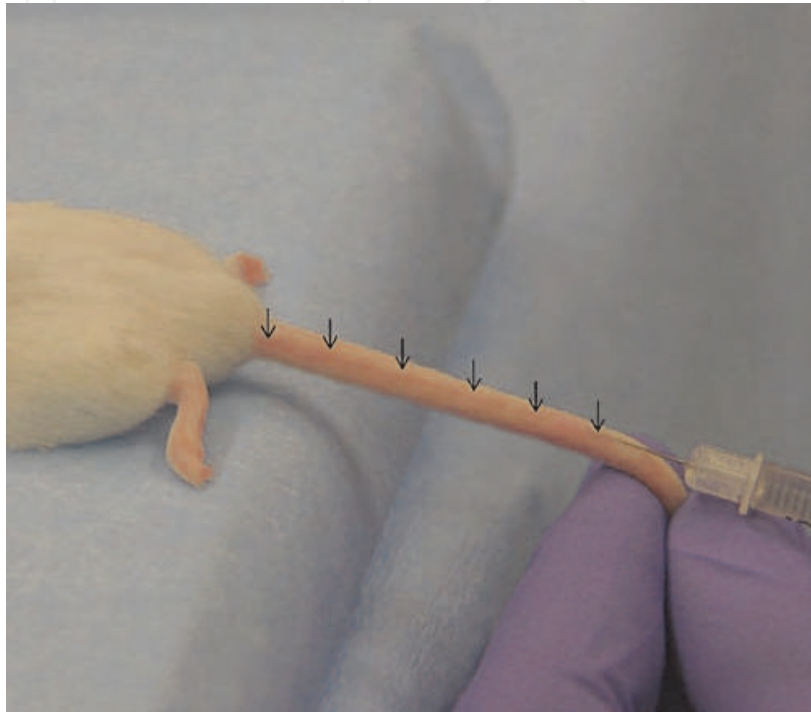


Photo 2. The tail is rotated 90° degrees and the needle is inserted in a shallow angle—arrows are depicting the dilated vein

For hydrodynamic gene delivery in mice muscle, under general anesthesia, a small latex tourniquet is applied above the knee and a small incision is made to expose the great saphenous vein. The infusion of the gene solution precedes the placement of a 30g catheter in the tail vein, and two minutes later the tourniquet is removed<sup>5,6</sup>.

### 3.2 Rat

The tail vein injection is the most common procedure in rats<sup>57-65</sup> as in mice with small modifications. For hydrodynamic infusion a needle or an over-the-needle intravenous catheter 23-27g can be used. The over-the-needle catheter insertion in the vein is followed by needle withdrawal and catheter advancement (photo 3). Blood entrance in the catheter confirms the correct placement of the catheter, secured by taping to the tail and connected with an extension to the infusion pump at a rate of 1-2 ml/sec<sup>54</sup>.

For selective infusion in rat's liver, the animal should be in general anesthesia and with the aid of a midline incision, the inferior vena cava (IVC) is isolated with 4-0 suture above and below the liver. At a midpoint, a 21-gauge needle is positioned while around the hole a 4-0 suture is secured to minimize leakage. Two minutes after the injection, the sutures above and below the IVC are removed and midline incision is closed with 4-0 absorbable suture<sup>7,8,9</sup>.



Photo 3. The vein is dilated with the aim of a tourniquet and an over-the-needle catheter is inserted.

A surgical protocol, modifying the above technique, has been developed by Inoue et al.<sup>10</sup> for selective infusion in liver. A 0.5mm silicon tube was advanced to the inferior vena cava (IVC) through the right common iliac vein for rapid injection of the solution, whereas the suprahepatic and the infrahepatic IVC were clamped. At the end of the procedure, clamps were removed and gently pressure was applied in the puncture point to minimize hemorrhage. Sawyer's group<sup>67</sup> verified Inoue results and calculated the optimum volume of DNA solution per g of liver to be 0.6 ml and the flow rate at 15 ml of solution per min per g liver for optimal hydrodynamic gene delivery to the liver.

A different approach for infusion in liver is to insert a 26g elastic cannula at the middle of the hepatic artery, through the gastroduodenal artery, and the celiac artery, the portal vein and the inferior vena cava are temporally clamped. Injection of the solution in the arterial vasculature is followed, fifteen second later, by clamps removal and gastroduodenal artery ligation<sup>11</sup>.

For hydrodynamic infusion in kidney, a 26-g catheter is inserted distally to clamped renal vein, at its origin, and blood flow is re-established immediately after the rapid infusion. Pressure should be applied for homeostasis and the abdomen is closed in standard technique<sup>68,12</sup>.

Hydrodynamic gene delivery in hind limb muscle is carried out through an inserted 28g needle catheter in the saphenous vein in direction to the knee, while a placed tourniquet, at a proximal part, forces the solution into the small veins ending up in the muscle tissue cells for 1-2 min post-injection<sup>13,14,15,16</sup>(photo 4). According to Danialou et al.<sup>17</sup> gene delivery in rat's muscle was applied by a 23 g butterfly needle positioned in the femoral artery, directed distally, and the obstruction of venous circulation was originated by a clamp on the femoral vein. Before the hydrodynamic infusion 2 mg of papaverin and 2 mg of histamine (1 mg/ml) in normal saline were injected and, 20 minutes following the infusion, the vascular clamp was removed.

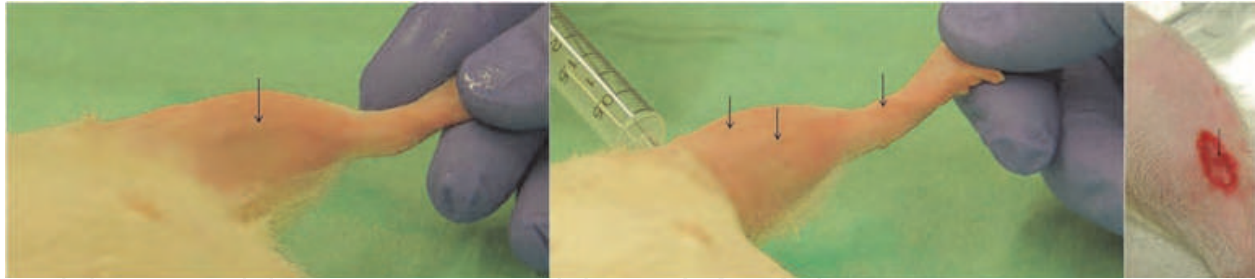


Photo 4. The saphenous vein prior to dilatation(left), after the placement of a tourniquet (middle), exposed after a small skin incision (right).

A protocol for hydrodynamic gene delivery in rat's heart was developed by de Carvalho and associates<sup>18</sup>. An incision at the fourth intercostal space exposed the heart and the aorta was identified, dissected from surrounding tissue and clamped. An injection, of 200  $\mu$ l PBS solution containing naked DNA in the left ventricle over 3 seconds, was followed, twenty seconds later, by clamp removal. During the procedure an ultrasound probe was attached toward the heart with an intensity of 1 MHz and power of 2 W/cm<sup>2</sup>

### 3.3 Rabbit

Eastman et al.<sup>19</sup>, in 2002, applied hydrodynamic gene delivery in larger animal model than rodents, avoiding heart failure that could be encountered when applied due to the large amount of infused solution. In anesthetized rabbit, in dorsal recumbent position, a right paramedian incision at the neck revealed the right jugular for the placement of a 20-g angiographic sheath and the insertion a 0.025 inch hydrophilic guide wire into a hepatic vein, under fluoroscopic control. The wire was replaced by a 5F balloon occlusion catheter in selected hepatic vein and inflated. A small amount of contrast material confirmed the proper position and the medium containing the macromolecule was infused rapidly at a rate 5 ml/sec at an 80 ml total volume. At the end of infusion the catheter was withdrawn and the incision was closed in standard technique.

The researchers modified this technique to block venous outflow during infusion. A balloon catheter was inserted in the hepatic vein, as described above, while from a second skin incision, over the femoral vein, a 20-g angiographic sheath was positioned and a 0.025 hydrophilic guide wire was inserted, through the sheath, into the proximal inferior vena cava (IVC) under fluoroscopic guidance (photo 5). A 7F introducer sheath substituted the angiographic sheath, reaching the distal IVC and enabling the introduction of a 5F XXL angioplasty catheter to the intrahepatic portion of the IVC to inhibit all venous outflow. After the infusion all catheters were removed.

Subsequently, a whole organ isolation protocol was demonstrated with the primary aid of a balloon catheter positioned, as already described through the jugular vein, above the upper hepatic veins. A second balloon catheter was inserted through the femoral vein between the lower hepatic veins and the renal veins, while, a third 4F pediatric pigtail infusion catheter was advanced from the contralateral femoral vein to a point between the two occlusion catheters. Vasovagal response was diminished by glycopyrrolate injection (0.01 mg/Kg) followed by the inflation of the catheters, and injection of a radiopaque solution, prior to the gene delivery, demonstrated the absence of a leakage. At the end of the surgical procedure all catheters were removed and bleeding was controlled.



Photo 5. The sheath is being inserted in the femoral vein of a rabbit.

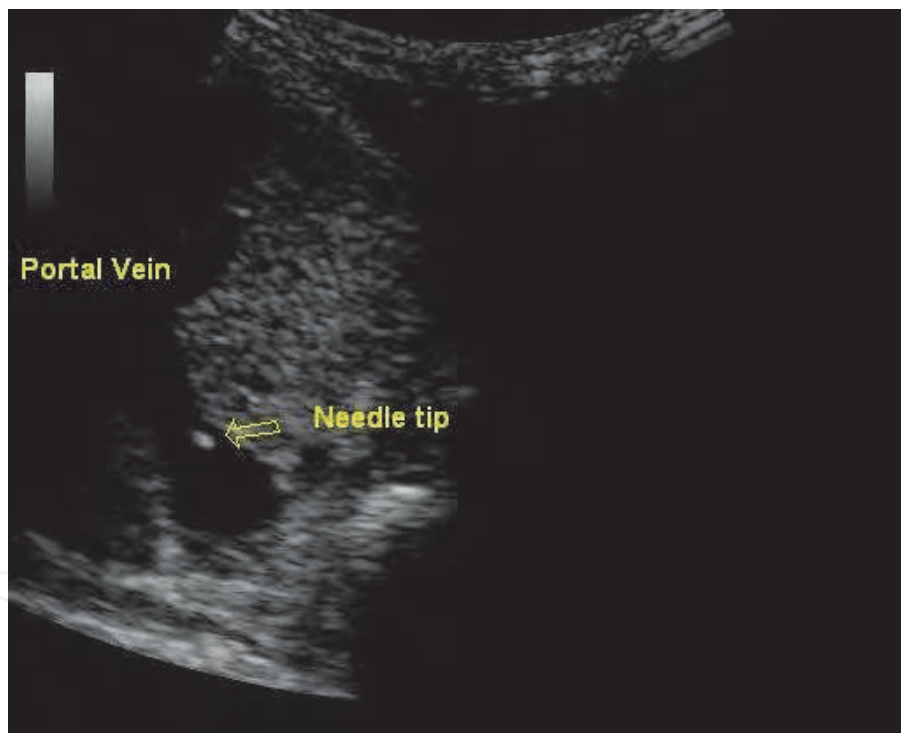


Photo 6. Insertion of a needle catheter in the portal vein, under ultrasound guidance.

The same group revealed the possibility to infuse percutaneously in the portal system, with a 20-g needle under ultrasound guidance, the gene solution ( photo 6). A small amount of contrast material was firstly delivered to confirm the correct placement of the needle in order a 0.025 glide wire to be advanced through the needle at liver parenchyma. An angiographic sheath was, then, placed over the wire and the position was verified again fluoroscopically. After the infusion, the catheter was removed and hemostasis was achieved with manual pressure at the insertion point.

### 3.4 Swine

Swine is the most popular large animal model for hydrodynamic gene delivery among the researcher groups the past years. Yoshino and associates<sup>20</sup> have developed a surgical protocol based on Eastman's research. Upon the insertion of an occlusion balloon catheter, through the right external jugular vein, into the left hepatic vein under fluoroscopic guidance, the hepatic artery and the portal vein were identified and clamped after a midline incision. The injection of 200 ml of saline, to wash out the liver blood through a catheter placed proximal to the clamp in the portal vein, was followed by the occlusion of the hepatic veins. The occlusion catheter was removed thirty seconds after the application of the hydrodynamic delivery, along with the clamps at the portal and hepatic veins, and the hepatic artery. The midline incision was closed in standard technique.

A modification of the technique above was introduced by Suda et al.<sup>68</sup> and Kamimura et al.<sup>21</sup>. Through an 18g catheter placed in the jugular vein, a 0.035 guide wire was inserted into the inferior vena cava (IVC) under fluoroscopic guidance. A 9F balloon catheter was advanced into the right lateral liver lobe over the guide wire, while two occlusions balloons 8F were inserted, one from the femoral vein into the IVC and the other one from the superior mesenteric vein into the portal trunk, to block leakage from the injected solution. Suda's group<sup>68</sup>, also, demonstrated a successful gene delivery in kidney with the aid of the balloon catheter inserted to the right renal vein, instead of the right liver lobe, and the IVC occlusion balloon, using the technique as already described.

Habib group<sup>22,23</sup> developed a minimal invasive surgical protocol to deliver gene in liver lobe's segment. After a right paramedian incision at the neck, the right external jugular vein was identified and isolated from the surrounding tissues and an introducer sheath was positioned in place. A hydrophilic catheter 5F was advanced, over a 0.035 guide wire previously positioned under fluoroscopy to the right hepatic vein, at the periphery of the right lateral liver lobe. A custom made 7F rigid balloon catheter, with multiple holes at the tip (photo 7), replaced the hydrophilic catheter remained in place for 10 minutes, after the infusion, to verify fluoroscopically the occlusion of the hepatic vein (photo 8). At the end of the procedure, the catheter was deflated, removed from the external jugular vein and a 3-0 absorbable ligature was used proximal to the puncture site to control bleeding.

Danielou et al.<sup>76</sup>, described a surgical protocol for gene delivery to hind limb muscle introducing an 8F catheter in the femoral artery directing distally, while a tourniquet was placed around the limb proximal to the catheter. 10 mg of papaverine was delivered in the arterial line over 30 sec, while femoral vein had been clamped, followed, 5 min later, by the injection of 10 mg of papaverine mixed with 10 mg of histamine. The gradual removal of the tourniquet, within 10 min from the gene infusion, was followed by catheter and clamp removal. The effects of histamine, if present, were counteracted by 50 mg of diphenhydramine and dextran volume expander. A different approach, for hydrodynamic delivery in muscle, has been described by Kamimura and associates<sup>24</sup>. Over a 0.035 guide wire, positioned to the femoral vein under fluoroscopic guidance through an 18g catheter in the jugular vein, an 8F balloon catheter is positioned. The isolated hind limb, by a rubber band proximally sited, remained in place until the infused balloon was slowly deflated 20 min after the infusion.

For hydrodynamic gene transfer to heart tissue, Alino et al.<sup>25</sup> placed a 7F Swan-Ganz catheter and a 6 F multipurpose catheter at the coronary sinus under fluoroscopic guidance,



through two 7F sheaths in the right and the left jugular veins, respectively. The coronary sinus vein was sealed by the Swan-Ganz catheter while the plasmid was injected through the multipurpose catheter and three minutes later the catheters were removed.

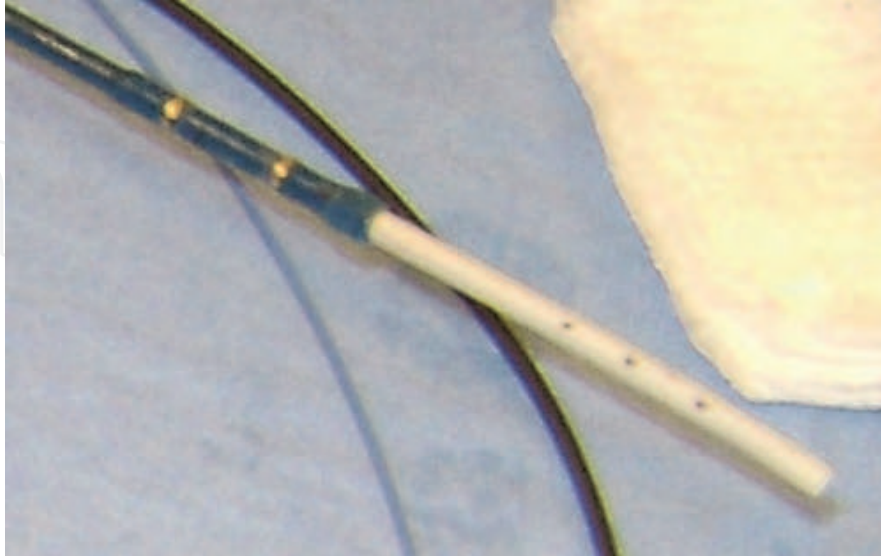


Photo 7. The custom made rigid-catheter with multiple holes at the side and a single at the center, enabling the infusion of large volumes of solution in small period of time without leakage.

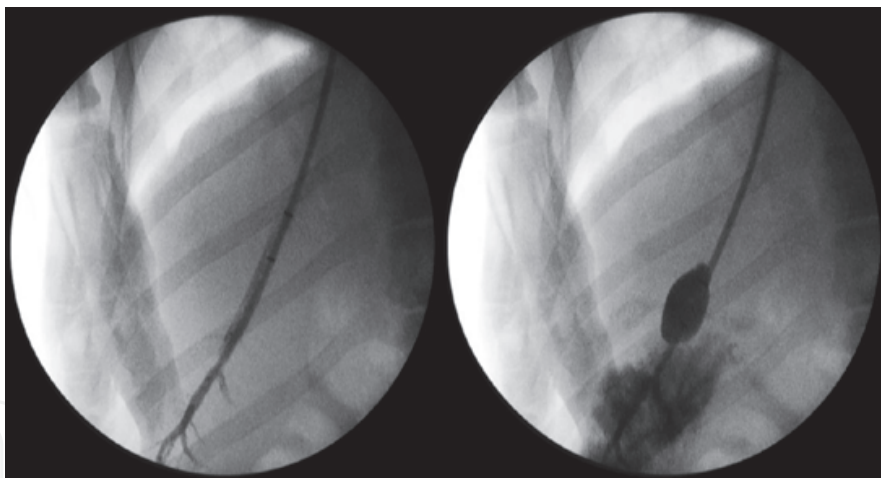


Photo 8. Placement of the catheter under fluoroscopic assistance (left), and the infused liver lobe with the balloon inflated without leakage of the gene solution and a small amount of contrast material (right).

### 3.5 Other animals - Human

#### 3.5.1 Canine

Canine, as an animal model, is limited, mainly, in muscle gene delivery. After sedation, a 20 g catheter is inserted in the great saphenous vein while a cuff has been positioned just above the stifle and inflated to 300 mm Hg pressure, to block blood flow. Before infusing the gene solution, 25ml of natural saline containing 4.2 mg of papaverine and 150 units of

heparin are delivered in 10sec, and five minutes later the solution containing the pDNA is injected at a rate of 2ml/sec<sup>72,75,26</sup>.

### 3.5.2 Primates

In nonhuman primates, Brunetti-Pierri's group<sup>27,28</sup> has established several different surgical protocols for gene delivery in liver. Saline solution (200ml) was firstly infused, followed by the vector injection in the portal vein, while its distal part and the hepatic artery had been occluded by vessel loops. The loops were retained in place for 30 minutes and the portal vein at the injection point was sutured by monofilament suture. In the second surgical protocol, the main strategy was to occlude not only the blood inflow but the outflow, as well. The surgical procedure described above was repeated and two umbilical tapes were passed around the infrahepatic IVC and suprahepatic IVC. 20ml/kg of normal saline were intravenously infused to prevent hypotension during total hepatic occlusion. Saline was again infused, at a volume of 12 to 13% of the total blood volume, through the portal vein catheter followed by the rapid occlusion of the infrahepatic and suprahepatic IVC and the hydrodynamic injection. In case of severe hypotension (mean arterial pressure less than 60 mm Hg) phenylephrine, at a dose of 40 mg in 250 ml of saline, was administrated. In the third protocol, a catheter was placed in the IVC to retrieve the unabsorbed vector modifying the previous protocol, while in the next protocol the retrieved vector was reinjected to the portal vein

A minimal invasive protocol was developed by the same research group. Two sheaths, 4F and 11 F, were positioned in the right and the left femoral vein, respectively, along with a 4F sheath in the left femoral artery. From the right femoral sheath, a custom made occlusion balloon-catheter was advanced in the IVC right arterial junction and deflated 7.5 to 15 min after the infusion from the catheter mounted in the hepatic artery, through the left femoral artery. Mild hypotension resolved with 20 ml/kg saline and phenylephrine to effect<sup>29</sup>.

In nonhuman primates, hydrodynamic delivery in muscle has also been well established. Under general anesthesia, after the placement of a 20g intravenous catheter into the distal cephalic vein, for the forearm, or into the small saphenous vein, for the hind limb, a tourniquet, above the elbow or the knee, is inflated to a pressure of 450 mmHg. The gene solution is rapidly infusion at a rate of 2 ml/sec and 2 minutes post-injection the tourniquet is deflated<sup>55,72,73,30,31,32</sup>.

### 3.5.3 Human

There is only one publication about hydrodynamic gene delivery in humans, by Khorsandi et al.<sup>82</sup>. A modified balloon catheter was introduced in the femoral vein, after the infusion of local anesthetic at the puncture site, and reached the middle hepatic vein, under fluoroscopy guidance. Before injecting the gene solution, the correct placement of the catheter and the absence of back flow were verified by injecting contrast material. Upon the completion of the infusion, the catheter remained in place for 10 minutes and then removed; the patients were discharged from hospital the same day.

## 4. Conclusion and future perspectives

Hydrodynamic delivery of macromolecules, membrane-impermeable, to cytoplasm is a physical method that gains popularity over the last years. The simplicity of the procedures

as already described, the lack of sophisticated equipment and the produced results, lead the hydrodynamic delivery to become a clinically feasible procedure in the near future. There is already one clinical trial on humans and we believe that the refinement of the procedure and the delivery in tissues other than liver or muscle, with minimal tissue damage and maximal delivery, will aim to new drug discovery and potential treatment of several human diseases.

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## 6. References

- [1] Liu D, Knapp JE. Hydrodynamics-based gene delivery. *Curr Opin Mol Ther.* 2001 Apr;3(2):192-7. [PubMed: 11338933]
- [2] Hagstrom JE. Plasmid-based gene delivery to target tissues in vivo: the intravascular approach. *Curr Opin Mol Ther.* 2003 Aug;5(4):338-44. [PubMed: 14513675]
- [3] Hodges BL, Scheule RK. Hydrodynamic delivery of DNA. *Expert Opin Biol Ther.* 2003 Sep;3(6):911-8. [PubMed: 12943450]
- [4] Kobayashi N, Nishikawa M, Takakura Y. The hydrodynamics-based procedure for controlling the pharmacokinetics of gene medicines at whole body, organ and cellular levels. *Adv Drug Deliv Rev.* 2005 Apr 5;57(5):713-31. [PubMed: 1575775]
- [5] Al-Dosari MS, Knapp JE, Liu D. Hydrodynamic delivery. *Adv Genet.* 2005;54:65-82. [PubMed: 16096008]
- [6] Gao X, Kim KS, Liu D. Nonviral gene delivery: what we know and what is next. *AAPS J.* 2007 Mar 23;9(1):E92-104. [PubMed: 17408239]
- [7] Herweijer H, Wolff JA. Gene therapy progress and prospects: hydrodynamic gene delivery. *Gene Ther.* 2007 Jan;14(2):99-107. [PubMed: 17167496]
- [8] Suda T, Liu D. Hydrodynamic gene delivery: its principles and applications. *Mol Ther.* 2007 Dec;15(12):2063-9. [PubMed: 17912237]
- [9] Lewis DL, Wolff JA. Systemic siRNA delivery via hydrodynamic intravascular injection. *Adv Drug Deliv Rev.* 2007 Mar 30;59(2-3):115-23. [PubMed: 17442446]
- [10] Bonamassa B, Hai L, Liu D. Hydrodynamic gene delivery and its applications in pharmaceutical research. *Pharm Res.* 2011 Apr;28(4):694-701. [PubMed: 21191634]
- [11] Zhang G, Gao X, Song YK, Vollmer R, Stolz DB, Gasiorowski JZ, Dean DA, Liu D. Hydroporation as the mechanism of hydrodynamic delivery. *Gene Ther.* 2004 Apr;11(8):675-82.
- [12] Kobayashi N, Kuramoto T, Yamaoka K, Hashida M, Takakura Y. Hepatic uptake and gene expression mechanisms following intravenous administration of plasmid DNA by conventional and hydrodynamics-based procedures. *J Pharmacol Exp Ther.* 2001 Jun;297(3):853-60.
- [13] Lecocq M, Andrianaivo F, Warnier MT, Wattiaux-De Coninck S, Wattiaux R, Jadot M. Uptake by mouse liver and intracellular fate of plasmid DNA after a rapid tail vein injection of a small or a large volume. *J Gene Med.* 2003 Feb;5(2):142-56.
- [14] Liu F, Song Y, Liu D. Hydrodynamics-based transfection in animals by systemic administration of plasmid DNA. *Gene Ther.* 1999 Jul;6(7):1258-66. [PubMed: 10455434]

- [15] Zhang G, Budker V, Wolff JA. High levels of foreign gene expression in hepatocytes after tail vein injections of naked plasmid DNA. *Hum Gene Ther.* 1999 Jul 1;10(10):1735-7. [PubMed: 10428218]
- [16] Jiang J, Yamato E, Miyazaki J. Intravenous delivery of naked plasmid DNA for in vivo cytokine expression. *Biochem Biophys Res Commun.* 2001 Dec 21;289(5):1088-92. [PubMed: 11741303]
- [17] Kitajima M, Tsuyama Y, Miyano-Kurosaki N, Takaku H. Anti-tumor effect of intravenous TNF $\alpha$  gene delivery naked plasmid DNA using a hydrodynamics-based procedure. *Nucleosides Nucleotides Nucleic Acids.* 2005;24(5-7):647-50. [PubMed: 16248005]
- [18] Arad U, Zeira E, El-Latif MA, Mukherjee S, Mitchell L, Pappo O, Galun E, Oppenheim A. Liver-targeted gene therapy by SV40-based vectors using the hydrodynamic injection method. *Hum Gene Ther.* 2005 Mar;16(3):361-71. [PubMed: 15812231]
- [19] Al-Dosari M, Zhang G, Knapp JE, Liu D. Direct assessment of promoter activity of human cytochrome p450 genes using optimized transfection in vitro and in vivo. *Biosci Rep.* 2006 Jun;26(3):217-29. [PubMed: 16850252]
- [20] Sebestyén MG, Budker VG, Budker T, Subbotin VM, Zhang G, Monahan SD, Lewis DL, Wong SC, Hagstrom JE, Wolff JA. Mechanism of plasmid delivery by hydrodynamic tail vein injection. I. Hepatocyte uptake of various molecules. *J Gene Med.* 2006 Jul;8(7):852-73. [PubMed: 16724360]
- [21] Budker VG, Subbotin VM, Budker T, Sebestyén MG, Zhang G, Wolff JA. Mechanism of plasmid delivery by hydrodynamic tail vein injection. II. Morphological studies. *J Gene Med.* 2006 Jul;8(7):874-88. [PubMed: 16718734]
- [22] Pergolizzi RG, Jin G, Chan D, Pierre L, Bussel J, Ferris B, Leopold PL, Crystal RG. Correction of a murine model of von Willebrand disease by gene transfer. *Blood.* 2006 Aug 1;108(3):862-9. [PubMed: 16638935]
- [23] Bell JB, Podetz-Pedersen KM, Aronovich EL, Belur LR, McIvor RS, Hackett PB. Preferential delivery of the Sleeping Beauty transposon system to livers of mice by hydrodynamic injection. *Nat Protoc.* 2007;2(12):3153-65 [PubMed: 18079715]
- [24] Fukushima M, Hattori Y, Tsukada H, Koga K, Kajiwara E, Kawano K, Kobayashi T, Kamata K, Maitani Y. Adiponectin gene therapy of streptozotocin-induced diabetic mice using hydrodynamic injection. *J Gene Med.* 2007 Nov;9(11):976-85. [PubMed: 17868184]
- [25] Dames P, Laner A, Maucksch C, Aneja MK, Rudolph C. Targeting of the glucocorticoid hormone receptor with plasmid DNA comprising glucocorticoid response elements improves nonviral gene transfer efficiency in the lungs of mice. *J Gene Med.* 2007 Sep;9(9):820-9. [PubMed: 17668918]
- [26] Hibbitt OC, Harbottle RP, Waddington SN, Bursill CA, Coutelle C, Channon KM, Wade-Martins R. Delivery and long-term expression of a 135 kb LDLR genomic DNA locus in vivo by hydrodynamic tail vein injection. *J Gene Med.* 2007 Jun;9(6):488-97. [PubMed: 17471590]
- [27] Aronovich EL, Bell JB, Belur LR, Gunther R, Koniar B, Erickson DC, Schachern PA, Matisse I, McIvor RS, Whitley CB, Hackett PB. Prolonged expression of a lysosomal enzyme in mouse liver after Sleeping Beauty transposon-mediated gene delivery: implications for non-viral gene therapy of mucopolysaccharidoses. *J Gene Med.* 2007 May;9(5):403-15. [PubMed: 17407189]
- [28] Rudolph C, Sieverling N, Schillinger U, Lesina E, Plank C, Thünemann AF, Schönberger H, Rosenecker J. Thyroid hormone (T3)-modification of polyethyleneglycol (PEG)-

- polyethyleneimine (PEI) graft copolymers for improved gene delivery to hepatocytes. *Biomaterials*. 2007 Apr;28(10):1900-11. [PubMed: 17196251]
- [29] Nguyen AT, Dow AC, Kupiec-Weglinski J, Busuttill RW, Lipshutz GS. Evaluation of gene promoters for liver expression by hydrodynamic gene transfer. *J Surg Res*. 2008 Jul;148(1):60-6. [PubMed: 18570932]
- [30] Jeong YS, Kim EJ, Shim CK, Hou JH, Kim JM, Choi HG, Kim WK, Oh YK. Modulation of biodistribution and expression of plasmid DNA following mesenchymal progenitor cell-based delivery. *J Drug Target*. 2008 Jun;16(5):405-14. [PubMed: 18569285]
- [31] Chilukuri N, Duysen EG, Parikh K, Sun W, Doctor BP, Lockridge O, Saxena A. Adenovirus-mediated gene transfer of human butyrylcholinesterase results in persistent high-level transgene expression in vivo. *Chem Biol Interact*. 2008 Sep 25;175(1-3):327-31. [PubMed: 18499092]
- [32] Li W, Ma N, Ong LL, Kaminski A, Skrabal C, Ugurlucan M, Lorenz P, Gatzen HH, Lützwow K, Lendlein A, Pützer BM, Li RK, Steinhoff G. Enhanced thoracic gene delivery by magnetic nanobead-mediated vector. *J Gene Med*. 2008 Aug;10(8):897-909. [PubMed: 18481827]
- [33] Wooddell CI, Reppen T, Wolff JA, Herweijer H. Sustained liver-specific transgene expression from the albumin promoter in mice following hydrodynamic plasmid DNA delivery. *J Gene Med*. 2008 May;10(5):551-63. [PubMed: 18330848]
- [34] Chen Q, Houry M, Chen J. Expression of human cytokines dramatically improves reconstitution of specific human-blood lineage cells in humanized mice. *Proc Natl Acad Sci U S A*. 2009 Dec 22;106(51):21783-8. [PubMed: 19966223]
- [35] Woodard LE, Keravala A, Jung WE, Wapinski OL, Yang Q, Felsher DW, Calos MP. Impact of hydrodynamic injection and phiC31 integrase on tumor latency in a mouse model of MYC-induced hepatocellular carcinoma. *PLoS One*. 2010 Jun 29;5(6):e11367. [PubMed: 20614008]
- [36] Belcher JD, Vineyard JV, Bruzzone CM, Chen C, Beckman JD, Nguyen J, Steer CJ, Vercellotti GM. Heme oxygenase-1 gene delivery by Sleeping Beauty inhibits vascular stasis in a murine model of sickle cell disease. *J Mol Med (Berl)*. 2010 Jul;88(7):665-75. [PubMed: 20306336]
- [37] Chen ZY, Liang K, Qiu RX. Targeted gene delivery in tumor xenografts by the combination of ultrasound-targeted microbubble destruction and polyethylenimine to inhibit surviving gene expression and induce apoptosis. *J Exp Clin Cancer Res*. 2010 Nov 23;29:152. [PubMed: 21092274]
- [38] Keng VW, Tschida BR, Bell JB, Largaespada DA. Modeling hepatitis B virus X-induced hepatocellular carcinoma in mice with the Sleeping Beauty transposon system. *Hepatology*. 2011 Mar;53(3):781-90. [PubMed: 21374658]
- [39] Cheng L, Wang J, Li X, Xing Q, Du P, Su L, Wang S. Interleukin-6 induces Gr-1+CD11b+ myeloid cells to suppress CD8+ T cell-mediated liver injury in mice. *PLoS One*. 2011 Mar 4;6(3):e17631. [PubMed: 21394214]
- [40] Miyakawa N, Nishikawa M, Takahashi Y, Ando M, Misaka M, Watanabe Y, Takakura Y. Prolonged circulation half-life of interferon  $\gamma$  activity by gene delivery of interferon  $\gamma$ -serum albumin fusion protein in mice. *J Pharm Sci*. 2011 Jun;100(6):2350-7. [PubMed: 21246562]
- [41] Schüttrumpf J, Milanov P, Abriss D, Roth S, Tonn T, Seifried E. Transgene loss and changes in the promoter methylation status as determinants for expression duration in nonviral gene transfer for factor IX. *Hum Gene Ther*. 2011 Jan;22(1):101-6. [PubMed: 20677911]

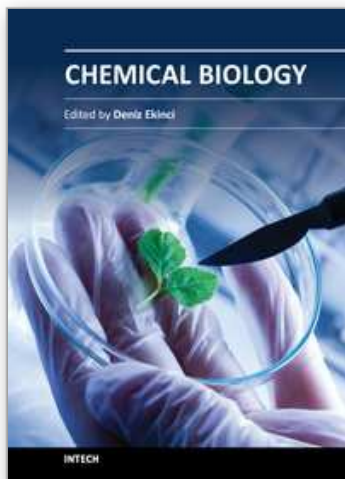
- [42] Kim HS, Kim JC, Lee YK, Kim JS, Park YS. Hepatic control elements promote long-term expression of human coagulation factor IX gene in hydrodynamically transfected mice. *J Gene Med.* 2011 Jul;13(7-8):365-72. [PubMed: 21710610]
- [43] Shahaf G, Moser H, Ozeri E, Mizrahi M, Abecassis A, Lewis EC.  $\alpha$ -1-Antitrypsin Gene Delivery Reduces Inflammation, Increases T-Regulatory Cell Population Size and Prevents Islet Allograft Rejection. *Mol Med.* 2011;17(9-10):1000-11. [PubMed: 21670848]
- [44] Doherty JE, Huye L, Yusa K, Zhou L, Craig N, Wilson M. Hyperactive piggyBac gene transfer in human cells and in vivo. *Hum Gene Ther.* 2011 Oct 12. [PubMed: 21992617]
- [45] Bu X, Zhou Y, Zhang H, Qiu W, Chen L, Cao H, Fang L, Wen P, Tan R, Yang J. Systemic administration of naked plasmid encoding HGF attenuates puromycin aminonucleoside-induced damage of murine glomerular podocytes. *Am J Physiol Renal Physiol.* 2011 Oct;301(4):F784-92. [PubMed: 21775482]
- [46] Abedini F, Ismail M, Hosseinkhani H, Ibrahim TA, Omar AR, Chong PP, Bejo MH, Domb AJ. Effects of CXCR4 siRNA/dextran-spermine nanoparticles on CXCR4 expression and serum LDH levels in a mouse model of colorectal cancer metastasis to the liver. *Cancer Manag Res.* 2011;3:301-9. [PubMed: 21931504]
- [47] Tripathi SK, Goyal R, Ansari KM, Ravi Ram K, Shukla Y, Chowdhuri DK, Gupta KC. Polyglutamic acid-based nanocomposites as efficient non-viral gene carriers in vitro and in vivo. *Eur J Pharm Biopharm.* 2011 Nov;79(3):473-84. [PubMed: 21820510]
- [48] Shi B, Keough E, Matter A, Leander K, Young S, Carlini E, Sachs AB, Tao W, Abrams M, Howell B, Sepp-Lorenzino L. Biodistribution of small interfering RNA at the organ and cellular levels after lipid nanoparticle-mediated delivery. *J Histochem Cytochem.* 2011 Aug;59(8):727-40. [PubMed: 21804077]
- [49] Hibbitt O, Wade-Martins R. High capacity extrachromosomal gene expression vectors. *Methods Mol Biol.* 2011;738:19-40. [PubMed: 21431717]
- [50] Keravala A, Chavez CL, Hu G, Woodard LE, Monahan PE, Calos MP. Long-term phenotypic correction in factor IX knockout mice by using phiC31 integrase-mediated gene therapy. *Gene Ther.* 2011 Aug;18(8):842-8. [PubMed: 21412285]
- [51] Parikh K, Duysen EG, Snow B, Jensen NS, Manne V, Lockridge O, Chilukuri N. Gene-delivered butyrylcholinesterase is prophylactic against the toxicity of chemical warfare nerve agents and organophosphorus compounds. *J Pharmacol Exp Ther.* 2011 Apr;337(1):92-101. [PubMed: 21205915]
- [52] Fernandez CA, Baumhover NJ, Duskey JT, Khargharia S, Kizzire K, Ericson MD, Rice KG. Metabolically stabilized long-circulating PEGylated polyacridine peptide polyplexes mediate hydrodynamically stimulated gene expression in liver. *Gene Ther.* 2011 Jan;18(1):23-37. [PubMed: 20720577]
- [53] Cao M, Khan JA, Kang BY, Mehta JL, Hermonat PL. Dual AAV/IL-10 Plus STAT3 Anti-Inflammatory Gene Delivery Lowers Atherosclerosis in LDLR KO Mice, but without Increased Benefit. *Int J Vasc Med.* 2012;2012:524235. [PubMed: 21915378]
- [54] Waynforth HB, Flecknell P. *Specific surgical operations. Experimental and Surgical Technique in the Rat.* 1992 Academic Press Ltd, London. 2nd ed, pp 44-48.
- [55] Wooddell CI, Subbotin VM, Sebestyén MG, Griffin JB, Zhang G, Schleef M, Braun S, Huss T, Wolff JA. Muscle damage after delivery of naked plasmid DNA into skeletal muscles is batch dependent. *Hum Gene Ther.* 2011 Feb;22(2):225-35. [PubMed: 20942645]

- [56] Itaka K, Osada K, Morii K, Kim P, Yun SH, Kataoka K. Polyplex nanomicelle promotes hydrodynamic gene introduction to skeletal muscle. *J Control Release*. 2010 Apr 2;143(1):112-9. [PubMed: 20043959]
- [57] Maruyama H, Higuchi N, Nishikawa Y, Kameda S, Iino N, Kazama JJ, Takahashi N, Sugawa M, Hanawa H, Tada N, Miyazaki J, Gejyo F. High-level expression of naked DNA delivered to rat liver via tail vein injection. *J Gene Med*. 2002 May-Jun;4(3):333-41. [PubMed: 12112650]
- [58] Higuchi N, Maruyama H, Kuroda T, Kameda S, Iino N, Kawachi H, Nishikawa Y, Hanawa H, Tahara H, Miyazaki J, Gejyo F. Hydrodynamics-based delivery of the viral interleukin-10 gene suppresses experimental crescentic glomerulonephritis in Wistar-Kyoto rats. *Gene Ther*. 2003 Aug;10(16):1297-310. [PubMed: 12883526]
- [59] Liu H, Hanawa H, Yoshida T, Elnaggar R, Hayashi M, Watanabe R, Toba K, Yoshida K, Chang H, Okura Y, Kato K, Kodama M, Maruyama H, Miyazaki J, Nakazawa M, Aizawa Y. Effect of hydrodynamics-based gene delivery of plasmid DNA encoding interleukin-1 receptor antagonist-Ig for treatment of rat autoimmune myocarditis: possible mechanism for lymphocytes and noncardiac cells. *Circulation*. 2005 Apr 5;111(13):1593-600. [PubMed: 15795329]
- [60] Elnaggar R, Hanawa H, Liu H, Yoshida T, Hayashi M, Watanabe R, Abe S, Toba K, Yoshida K, Chang H, Minagawa S, Okura Y, Kato K, Kodama M, Maruyama H, Miyazaki J, Aizawa Y. The effect of hydrodynamics-based delivery of an IL-13-Ig fusion gene for experimental autoimmune myocarditis in rats and its possible mechanism. *Eur J Immunol*. 2005 Jun;35(6):1995-2005. [PubMed: 15902684]
- [61] Abe S, Hanawa H, Hayashi M, Yoshida T, Komura S, Watanabe R, Lie H, Chang H, Kato K, Kodama M, Maruyama H, Nakazawa M, Miyazaki J, Aizawa Y. Prevention of experimental autoimmune myocarditis by hydrodynamics-based naked plasmid DNA encoding CTLA4-Ig gene delivery. *J Card Fail*. 2005 Sep;11(7):557-64. [PubMed: 16198253]
- [62] Toietta G, Mane VP, Norona WS, Finegold MJ, Ng P, McDonagh AF, Beaudet AL, Lee B. Lifelong elimination of hyperbilirubinemia in the Gunn rat with a single injection of helper-dependent adenoviral vector. *Proc Natl Acad Sci USA*. 2005 Mar 15;102(11):3930-5. [PubMed: 15753292]
- [63] Tada M, Hatano E, Taura K, Nitta T, Koizumi N, Ikai I, Shimahara Y. High volume hydrodynamic injection of plasmid DNA via the hepatic artery results in a high level of gene expression in rat hepatocellular carcinoma induced by diethylnitrosamine. *J Gene Med*. 2006 Aug;8(8):1018-26. [PubMed: 16779866]
- [64] Takekubo M, Tsuchida M, Haga M, Saitoh M, Hanawa H, Maruyama H, Miyazaki J, Hayashi J. Hydrodynamics-based delivery of plasmid DNA encoding CTLA4-Ig prolonged cardiac allograft survival in rats. *J Gene Med*. 2008 Mar;10(3):290-7. [PubMed: 18074399]
- [65] Dimmock D, Brunetti-Pierri N, Palmer DJ, Beaudet AL, Ng P. Correction of hyperbilirubinemia in gunn rats using clinically relevant low doses of helper-dependent adenoviral vectors. *Hum Gene Ther*. 2011 Apr;22(4):483-8. [PubMed: 20973621]
- [66] Salehi S, Eckley L, Sawyer GJ, Zhang X, Dong X, Freund JN, Fabre JW. Intestinal lactase as an autologous beta-galactosidase reporter gene for in vivo gene expression studies. *Hum Gene Ther*. 2009 Jan;20(1):21-30. [PubMed: 20377368]
- [67] Sawyer GJ, Grehan A, Dong X, Whitehorne M, Seddon M, Shah AM, Zhang X, Salehi S, Fabre JW. Low-volume hydrodynamic gene delivery to the rat liver via an isolated

- segment of the inferior vena cava: efficiency, cardiovascular response and intrahepatic vascular dynamics. *J Gene Med.* 2008 May;10(5):540-50. [PubMed: 18307279]
- [68] Suda T, Suda K, Liu D. Computer-assisted hydrodynamic gene delivery. *Mol Ther.* 2008 Jun;16(6):1098-104. [PubMed: 18398428]
- [69] Inoue S, Hakamata Y, Kaneko M, Kobayashi E. Gene therapy for organ grafts using rapid injection of naked DNA: application to the rat liver. *Transplantation.* 2004 Apr 15;77(7):997-1003. [PubMed: 15087760]
- [70] Tada M, Hatano E, Taura K, Nitta T, Koizumi N, Ikai I, Shimahara Y. High volume hydrodynamic injection of plasmid DNA via the hepatic artery results in a high level of gene expression in rat hepatocellular carcinoma induced by diethylnitrosamine. *J Gene Med.* 2006 Aug;8(8):1018-26. [PubMed: 16779866]
- [71] Xing Y, Pua EC, Lu X, Zhong P. Low-amplitude ultrasound enhances hydrodynamic-based gene delivery to rat kidney. *Biochem Biophys Res Commun.* 2009 Aug 14;386(1):217-22. [PubMed: 19523454]
- [72] Hagstrom JE, Hegge J, Zhang G, Noble M, Budker V, Lewis DL, Herweijer H, Wolff JA. A facile nonviral method for delivering genes and siRNAs to skeletal muscle of mammalian limbs. *Mol Ther.* 2004 Aug;10(2):386-98. [PubMed: 15294185]
- [73] Sebestyén MG, Hegge JO, Noble MA, Lewis DL, Herweijer H, Wolff JA. Progress toward a nonviral gene therapy protocol for the treatment of anemia. *Hum Gene Ther.* 2007 Mar;18(3):269-85. [PubMed: 17376007]
- [74] Shi Q, Wang H, Tran C, Qiu X, Winnik FM, Zhang X, Dai K, Benderdour M, Fernandes JC. Hydrodynamic delivery of chitosan-folate-DNA nanoparticles in rats with adjuvant-induced arthritis. *J Biomed Biotechnol.* 2011;2011:148763. [PubMed: 21274258]
- [75] Su LT, Gopal K, Wang Z, Yin X, Nelson A, Kozyak BW, Burkman JM, Mitchell MA, Low DW, Bridges CR, Stedman HH. Uniform scale-independent gene transfer to striated muscle after transvenular extravasation of vector. *Circulation.* 2005 Sep 20;112(12):1780-8. [PubMed: 16157771]
- [76] Danialou G, Comtois AS, Matecki S, Nalbantoglu J, Karpati G, Gilbert R, Geoffroy P, Gilligan S, Tanguay JF, Petrof BJ. Optimization of regional intraarterial naked DNA-mediated transgene delivery to skeletal muscles in a large animal model. *Mol Ther.* 2005 Feb;11(2):257-66. [PubMed: 15668137]
- [77] Pinto de Carvalho L, Takeshita D, Carillo BA, Garcia Lisboa BC, Molina G, Beutel A, Yasumura EG, Takiya CM, Valero VB, Ribeiro de Campos R Jr, Dohmann HF, Han SW. Hydrodynamics- and ultrasound-based transfection of heart with naked plasmid DNA. *Hum Gene Ther.* 2007 Dec;18(12):1233-43. [PubMed: 18021018]
- [78] Eastman SJ, Baskin KM, Hodges BL, Chu Q, Gates A, Dreusicke R, Anderson S, Scheule RK. Development of catheter-based procedures for transducing the isolated rabbit liver with plasmid DNA. *Hum Gene Ther.* 2002 Nov 20;13(17):2065-77. [PubMed: 12490001]
- [79] Yoshino H, Hashizume K, Kobayashi E. Naked plasmid DNA transfer to the porcine liver using rapid injection with large volume. *Gene Ther.* 2006 Dec;13(24):1696-702. [PubMed: 16871229]
- [80] Kamimura K, Suda T, Xu W, Zhang G, Liu D. Image-guided, lobe-specific hydrodynamic gene delivery to swine liver. *Mol Ther.* 2009 Mar;17(3):491-9. [PubMed: 19156134]
- [81] Katsimpoulas M, Zacharoulis D, Rountas C, Dimitriou C, Mantziaras G, Kostomitsopoulos N, Habib N, Kostakis A. Minimal invasive technique for gene delivery in porcine liver lobe segment. *J Invest Surg.* 2011;24(1):13-7.



- [82] Khorsandi SE, Bachellier P, Weber JC, Greget M, Jaeck D, Zacharoulis D, Rountas C, Helmy S, Helmy A, Al-Waracky M, Salama H, Jiao L, Nicholls J, Davies AJ, Levicar N, Jensen S, Habib N. Minimally invasive and selective hydrodynamic gene therapy of liver segments in the pig and human. *Cancer Gene Ther.* 2008 Apr;15(4):225-30.
- [83] Kamimura K, Zhang G, Liu D. Image-guided, intravascular hydrodynamic gene delivery to skeletal muscle in pigs. *Mol Ther.* 2010 Jan;18(1):93-100.
- [84] Aliño SF, José Herrero M, Bodi V, Noguera I, Mainar L, Dasí F, Sempere A, Sánchez M, Díaz A, Sabater L, Lledó S. Naked DNA delivery to whole pig cardiac tissue by coronary sinus retrograde injection employing non-invasive catheterization. *J Gene Med.* 2010 Nov;12(11):920-6.
- [85] Qiao C, Li J, Zheng H, Bogan J, Li J, Yuan Z, Zhang C, Bogan D, Kornegay J, Xiao X. Hydrodynamic limb vein injection of adeno-associated virus serotype 8 vector carrying canine myostatin propeptide gene into normal dogs enhances muscle growth. *Hum Gene Ther.* 2009 Jan;20(1):1-10. [PubMed: 18828709]
- [86] Brunetti-Pierri N, Ng T, Iannitti DA, Palmer DJ, Beaudet AL, Finegold MJ, Carey KD, Cioffi WG, Ng P. Improved hepatic transduction, reduced systemic vector dissemination, and long-term transgene expression by delivering helper-dependent adenoviral vectors into the surgically isolated liver of nonhuman primates. *Hum Gene Ther.* 2006 Apr;17(4):391-404. [PubMed: 12490001]
- [87] Brunetti-Pierri N, Ng P. Progress towards the clinical application of helper-dependent adenoviral vectors for liver and lung gene therapy. *Curr Opin Mol Ther.* 2006 Oct;8(5):446-54. [PubMed: 17078387]
- [88] Brunetti-Pierri N, Stapleton GE, Law M, Breinholt J, Palmer DJ, Zuo Y, Grove NC, Finegold MJ, Rice K, Beaudet AL, Mullins CE, Ng P. Efficient, long-term hepatic gene transfer using clinically relevant HDAd doses by balloon occlusion catheter delivery in nonhuman primates. *Mol Ther.* 2009 Feb;17(2):327-33. [PubMed: 19050700]
- [89] Zhang G, Budker V, Williams P, Subbotin V, Wolff JA. Efficient expression of naked dna delivered intraarterially to limb muscles of nonhuman primates. *Hum Gene Ther.* 2001 Mar 1;12(4):427-38. [PubMed: 11242534]
- [90] Vigen KK, Hegge JO, Zhang G, Mukherjee R, Braun S, Grist TM, Wolff JA. Magnetic resonance imaging-monitored plasmid DNA delivery in primate limb muscle. *Hum Gene Ther.* 2007 Mar;18(3):257-68. [PubMed: 17376006]
- [91] Toromanoff A, Chérel Y, Guilbaud M, Penaud-Budloo M, Snyder RO, Haskins ME, Deschamps JY, Guigand L, Podevin G, Arruda VR, High KA, Stedman HH, Rolling F, Anegón I, Moullier P, Le Guiner C. Safety and efficacy of regional intravenous (r.i.) versus intramuscular (i.m.) delivery of rAAV1 and rAAV8 to nonhuman primate skeletal muscle. *Mol Ther.* 2008 Jul;16(7):1291-9. [PubMed: 18461055]



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Chemical biology utilizes chemical principles to modulate systems to either investigate the underlying biology or create new function. Over recent years, chemical biology has received particular attention of many scientists in the life sciences from botany to medicine. This book contains an overview focusing on the research area of protein purification, enzymology, vitamins, antioxidants, biotransformation, gene delivery, signaling, regulation and organization. Particular emphasis is devoted to both theoretical and experimental aspects. The textbook is written by international scientists with expertise in synthetic chemistry, protein biochemistry, enzymology, molecular biology, drug discovery and genetics many of which are active chemical, biochemical and biomedical research. The textbook is expected to enhance the knowledge of scientists in the complexities of chemical and biological approaches and stimulate both professionals and students to dedicate part of their future research in understanding relevant mechanisms and applications of chemical biology.

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