CLINIGENE CURRENT GENE THERAPY WEEKLY

From April 18th to April 25th 2011

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PMID: 21512508

Gene Ther. 2011 Apr 21. [Epub ahead of print]

Tumor growth and metastasis suppression by Glipr1 gene-modified macrophages in a metastatic prostate cancer model.

Tabata K, Kurosaka S, Watanabe M, Edamura K, Satoh T, Yang G, Abdelfattah E, Wang J, Goltsov A, Floryk D, Thompson TC.

Scott Department of Urology, Baylor College of Medicine, Houston, TX, USA.

We previously identified the mouse and human Glipr1 and GLIPR1/RTVP-1 genes, respectively, as direct p53 targets with proapoptotic activities in various cancer cell lines, including prostate cancer (PCa). Intratumoral injection of an adenoviral vector capable of efficient transduction and expression of Glipr1 (AdGlipr1) yielded promising therapeutic results in an orthotopic, metastatic mouse model of PCa. AdGlipr1-transduced macrophages (Mφ/Glipr1) generated greater surface expression of CD40, CD80 and major histocompatibility complex class II molecules and greater production of interleukin 12 (IL-12) and IL-6 in vitro than control macrophages did. Mechanistic analysis indicated that increased production of IL-12 in Mφ/Glipr1 depends on activation of the p38 signaling cascade. Mφ/Glipr1 injected into orthotopic 178-2BMA tumors in vivo resulted in significantly suppressed prostate tumor growth and spontaneous lung metastases and longer survival relative to those observed in control-treated mice. Furthermore, these preclinical data indicate the generation of systemic natural killer cell activity and tumor-specific cytotoxic T lymphocyte responses. Trafficking studies confirmed that intratumorally injected Mφ/Glipr1 could migrate to draining lymph nodes. Overall, our data suggest that this novel gene-modified cell approach is an effective treatment avenue that induces antitumor immune responses in preclinical studies.

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Gene Ther. 2011 Apr 21. [Epub ahead of print]

Adeno-associated virus serotype 9-mediated pulmonary transgene expression: effect of mouse strain, animal gender and lung inflammation.

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Gene therapy holds great potential for the treatment of various acquired and inherited pulmonary diseases. Among various viral vectors, adeno-associated viral (AAV) vectors have been most frequently used in different clinical trials of pulmonary gene therapy. In the present study, we examined the kinetics and duration of transgene expression, vector biodistribution and development of neutralizing antibodies (NAB) in mice after pulmonary application of AAV2/9 vector. The pulmonary route of application did not affect any of the measured parameters. Transgene expression and biodistribution analysis at day 450 post-application confirmed the systemic spread of the vector after pulmonary delivery. Using SPB(-/-) mice, the study shows that AAV2/9-mediated gene expression is influenced by animal gender but not mouse genotype and is insensitive to the presence of lung inflammation. Lower expression levels were observed in male compared with female mice, and transient immunosuppression with dexamethasone significantly reduced the development of NAB in both genders of mice. The study thus advances this serotype for further development and use as a therapeutic vector.

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Gene Ther. 2011 Apr 21. [Epub ahead of print]

Delivery of gelfoam-enabled cells and vectors into the pericardial space using a percutaneous approach in a porcine model.

Ladage D, Turnbull IC, Ishikawa K, Takewa Y, Rapti K, Morel C, Karakikes I, Hadri L, Müller-Ehmsen J, Costa KD, Hajjar RJ, Kawase Y.
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Intrapericardial drug delivery is a promising procedure, with the ability to localize therapeutics with the heart. Gelfoam particles are nontoxic, inexpensive, nonimmunogenic and biodegradable compounds that can be used to deliver therapeutic agents. We developed a new percutaneous approach method for intrapericardial injection, puncturing the pericardial sac safely under fluoroscopy and intravascular ultrasound (IVUS) guidance. In a porcine model of myocardial infarction (MI), we deployed gelfoam particles carrying either (a) autologous mesenchymal stem cells (MSCs) or (b) an adenovirus encoding enhanced green fluorescent protein (eGFP) 48 h post-MI. The presence of MSCs and viral infection at the infarct zone was confirmed by immunoflourescence and PCR. Puncture was performed successfully in 16 animals. Using IVUS, we successfully determined the size of the pericardial space before the puncture, and safely accessed that space in setting of pericardial effusion and also adhesions induced by the MI. Intrapericardial injection of gelfoam was safe and reliable. Presence of the MSCs and eGFP expression from adenovirus in the myocardium were confirmed after delivery. Our novel percutaneous approach to deliver (stem-) cells or adenovirus was safe and efficient in this pre-clinical model. IVUS-guided delivery is a minimally invasive procedure that seems to be a promising new strategy to deliver therapeutic agents locally to the heart.

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Gene Ther. 2011 Apr 21. [Epub ahead of print]

Pre-clinical evaluation of three non-viral gene transfer agents for cystic fibrosis after aerosol delivery to the ovine lung.

McLachlan G, Davidson H, Holder E, Davies LA, Pringle IA, Sumner-Jones SG, Baker A, Tennant P, Gordon C, Vrettou C, Blundell R, Hyndman L, Stevenson B, Wilson A, Doherty A, Shaw DJ, Coles RL, Painter H, Cheng SH, Scheule RK, Davies JC, Innes JA, Hyde SC, Griesenbach U, Alton EW, Boyd AC, Porteous DJ, Gill DR, Collie DD.
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We use both large and small animal models in our pre-clinical evaluation of gene transfer agents (GTAs) for cystic fibrosis (CF) gene therapy. Here, we report the use of a large animal model to assess three non-viral GTAs: 25 kDa-branched polyethyleneimine (PEI), the cationic liposome (GL67A) and compacted DNA nanoparticle formulated with polyethylene glycol-substituted lysine 30-mer. GTAs complexed with plasmids expressing human cystic fibrosis transmembrane conductance regulator (CFTR) complementary DNA were administered to the sheep lung (n=8 per group) by aerosol. All GTAs gave evidence of gene transfer and expression 1 day after treatment. Vector-derived mRNA was expressed in lung tissues, including epithelial cell-enriched bronchial brushing samples, with median group values reaching 1-10% of endogenous CFTR mRNA levels. GL67A gave the highest levels of expression. Human CFTR protein was detected in small airway epithelial cells in some animals treated with GL67A (two out of eight) and PEI (one out of eight). Bronchoalveolar lavage neutrophilia, lung histology and elevated serum haptoglobin levels indicated that gene delivery was associated with mild local and systemic inflammation. Our conclusion was that GL67A was the best non-viral GTA currently available for aerosol delivery to the sheep lung, led to the selection of GL67A as our lead GTA for clinical trials in CF patients.

PMID: 21511296

J Urol. 2011 Apr 19. [Epub ahead of print]

Brain-Specific Angiogenesis Inhibitor 1 is a Putative Factor for Inhibition of Neovascular Formation in Renal Cell Carcinoma.

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PURPOSE:
Renal cell carcinoma is a typical hypervascular tumor in which neovascularization may have a large part in progression. We examined expression of the cancer regulating, p53 targeted angiogenesis inhibitor brain-specific angiogenesis inhibitor 1 in renal cell carcinoma tissue to elucidate the clinical significance of its expression.

MATERIALS AND METHODS:
We examined brain-specific angiogenesis inhibitor 1 mRNA and protein expression in 47 renal cell carcinoma and 10 normal kidney tissues using real-time quantitative polymerase chain reaction and immunohistochemistry, respectively. Levels of VEGF and bFGF mRNA, and immunohistochemical expression of p53 protein were also investigated in the same renal cell carcinoma tissues.

RESULTS:
A significant decrease in BAI1 mRNA was noted in renal cell carcinoma tissue compared with that in normal kidney tissue ($p < 0.001$). Immunostaining for brain-specific angiogenesis inhibitor 1 was also decreased in carcinoma tissue compared with normal kidney tissue. BAI1 mRNA and protein expression were lower in advanced renal cell carcinoma (pT3-4) than in localized renal cell carcinoma (pT1-2) tissues ($p < 0.03$ and 0.003, respectively). A significant negative correlation was observed between microvessel density and brain-specific angiogenesis inhibitor 1 protein expression ($r = -0.4056$, $p = 0.002$). No significant correlation was noted between BAI1 and VEGF or bFGF mRNA levels. Brain-specific angiogenesis inhibitor 1 protein expression did not correlate with p53 protein expression.

CONCLUSIONS:
These observations suggest that down-regulation of brain-specific angiogenesis inhibitor 1 expression may be a critical factor in renal cell carcinoma development and BAI1 may be a promising candidate for gene therapy of renal cell carcinoma.

PMID: 21510919

Biochem Biophys Res Commun. 2011 Apr 12. [Epub ahead of print]

Functional and physical competition between phospholamban and its mutants provides insight into the molecular mechanism of gene therapy for heart failure.

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We have used functional co-reconstitution of purified sarcoplasmic reticulum (SR) Ca(2+)-ATPase (SERCA) with phospholamban (PLB), its inhibitor in the heart, to test the hypothesis that loss-of-function (LOF) PLB mutants (PLB(M)) can compete with wild-type PLB (PLB(W)) to relieve SERCA inhibition. Co-reconstitution at varying PLB-to-SERCA ratios was conducted using synthetic PLB(W), gain-of-function mutant I40A, or LOF mutants S16E (phosphorylation mimic) or L31A. Inhibitory potency was defined as the fractional increase in K(Ca), measured from the Ca(2+)-dependence of ATPase activity. At saturating PLB, the inhibitory potency of I40A was about three times that of PLB(W), while the potency of each of the LOF PLB(M) was about one third that of PLB(W). However, there was no significant variation in the apparent SERCA affinity for these four PLB variants. When SERCA was co-reconstituted with mixtures of PLB(W) and LOF PLB(M), inhibitory potency was reduced relative to that of PLB(W) alone. Furthermore, FRET between donor-labeled SERCA and acceptor-labeled PLB(W) was decreased by both (unlabeled) LOF PLB(M). These results show that LOF PLB(M) can compete both physically and functionally with PLB(W), provide a rational explanation for the partial success of S16E-based gene therapy in animal models of heart failure, and establish a powerful platform for designing and testing more effective PLB(M) targeted for gene therapy of heart failure in humans.

PMID: 21510701

Mol Pharm. 2011 Apr 29. [Epub ahead of print]

Anionic Liposomes Enhance and Prolong Adenovirus-Mediated Gene Expression in Airway Epithelia in Vitro and in Vivo.

Zhong Z, Han J, Wan Y, Zhang Z, Sun X.
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Abstract
Adenoviral vector mediated gene therapy has received extensive attention in airway disease treatment. However, the lack of the requisite coxsackie-adenovirus receptor (CAR) on the apical surface of airway epithelium and the host immune response to adenoviruses limit their in vivo application. In our study, we developed for the first time a novel formulation composed of anionic liposomes and adenoviruses (AL-Ad5) using a calcium-induced phase change method. The obtained formulation was employed to enhance the transduction efficiency of airway gene delivery. Our results indicated that primary cultured airway epithelial cells infected by AL-Ad5 displayed higher LacZ gene expression compared to naked adenovirus. Importantly, AL-Ad5 significantly improved and prolonged LacZ gene expression in murine airway tissues when delivered in vivo by intratracheal instillation. Additionally, it was found that anionic liposomes provided immunoprotection to the adenovirus from neutralizing antibody, thus slowing down the elimination of Ad5 particles meanwhile reducing the inflammatory reaction caused by the Ad5 vector. These results suggested that the combination of anionic liposomes with adenovirus may be a useful strategy to deliver therapeutic genes into the airway epithelia and is promising in clinical application.

PMID: 21509783

Int J Cancer. 2011 Jan 20. doi: 10.1002/ijc.25948. [Epub ahead of print]

Combining conditionally replicating adenovirus-mediated gene therapy with chemotherapy: A novel antitumor approach.

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Despite significant improvements in diagnosis and innovations in the therapy of specific cancers, effective treatment of neoplastic diseases still presents major challenges. Recent studies have shown that conditionally replicating adenoviruses (CRAds) not only have the ability to destroy cancer cells but may also be potential vectors for the expression of therapeutic genes. Several studies in animal models have demonstrated that the combination of CRAds-mediated gene therapy and chemotherapy has greater therapeutic benefit than either treatment modality alone. In this review, an overview of specifications for a novel antitumor approach combining CRAd-gene therapy and chemotherapy is provided and recent progress in this field is discussed.

PMID: 21508376

Anticancer Res. 2011 Apr;31(4):1279-87.

Adenoviral Therapy Is More Effective in Gemcitabine-resistant Pancreatic Cancer than in Gemcitabine-sensitive Cells.

Yasui T, Ohuchida K, Zhao M, Cui L, Onimaru M, Egami T, Fujita H, Ohtsuka T, Mizumoto K, Matsumoto K, Tanaka M.
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BACKGROUND:
Although gemcitabine is the standard treatment for pancreatic cancer, this particular type of cancer develops rapidly and has intrinsic chemoresistance. Chemoresistance plays a critical role in tumor progression, invasion and migration. Nevertheless, the effect of adenoviral therapy on chemoresistant cancer cells has not been studied. In this study, we compared the efficacy of adenoviral therapy in parental and chemoresistant pancreatic cancer cells.

MATERIALS AND METHODS:
To establish gemcitabine-resistant cells, pancreatic cancer SUIT2 cells were exposed to increasing concentrations of gemcitabine. Both parental and chemoresistant cells were infected with adenoviruses expressing either green fluorescent protein (Ad-GFP) or the hepatocyte growth factor antagonist, NK4 (Ad-NK4). To investigate the transduction efficacy, GFP expression and NK4 concentrations were measured and an invasion assay was used to investigate the efficacy of the adenoviral therapy.

RESULTS:
The 50% inhibitory concentration of gemcitabine was <10 nM in the parental SUIT-2 cells, while it was >1 μM in gemcitabine-resistant cells. A large number of gemcitabine-resistant cells were GFP-positive compared with only a small number of parental cells ($p<0.05$). The NK4 expression level was significantly higher in gemcitabine-resistant cells than in parental cells ($p<0.05$). The supernatant from Ad-NK4-infected gemcitabine-resistant cells significantly inhibited the invasion of cancer cells compared with that from Ad-NK4-infected parental cells ($p<0.05$).
CONCLUSION:
Both the efficiency of transduction and the therapeutic efficacy of adenoviral therapy were higher in gemcitabine-resistant cells than in parental cells, suggesting that adenoviral gene therapy is more effective in patients with gemcitabine-resistant pancreatic cancer.

PMID: 21505426

Mol Ther. 2011 Apr 19. [Epub ahead of print]

Combined Flt3L/TK Gene Therapy Induces Immunological Surveillance Which Mediates an Immune Response Against a Surrogate Brain Tumor Neoantigen.

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Glioblastoma multiforme (GBM) is a primary brain tumor with a median survival of 14.6 months postdiagnosis. The infiltrative nature of GBM prevents complete resection and residual brain tumor cells give rise to recurrent GBM, a hallmark of this disease. Recurrent GBMs are known to harbor numerous mutations/gene rearrangements when compared to the primary tumor, which leads to the potential expression of novel proteins that could serve as tumor neoantigens. We have developed a combined immune-based gene therapeutic approach for GBM using adenoviral (Ads) mediated gene delivery of Herpes Simplex Virus Type 1-thymidine kinase (TK) into the tumor mass to induce tumor cells' death combined with an adenovirus expressing fms-like tyrosine kinase 3 ligand (Flt3L) to recruit dendritic cells (DCs) into the tumor microenvironment. This leads to the induction of specific anti-brain tumor immunity and immunological memory. In a model of GBM recurrence, we demonstrate that Flt3L/TK mediated immunological memory is capable of recognizing brain tumor neoantigens absent from the original treated tumor. These data demonstrate that the Flt3L/TK gene therapeutic approach can induce systemic immunological memory capable of recognizing a brain tumor neoantigen in a model of recurrent GBM.

PMID: 21505069

Hum Mol Genet. 2011 May 2. [Epub ahead of print]

Ex vivo gene therapy for HIV-1 treatment.

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Until recently, progress in ex vivo gene therapy (GT) for human immunodeficiency virus-1 (HIV-1) treatment has been incremental. Long-term HIV-1 remission in a patient who received a heterologous stem cell transplant for acquired immunodeficiency syndrome-related lymphoma from a CCR5(-/-) donor, even after discontinuation of conventional therapy, has energized the field. We review the status of current approaches as well as future directions in the areas of therapeutic targets, combinatorial strategies, vector design, introduction of therapeutics into stem cells and enrichment/expansion of gene-modified cells. Finally, we discuss recent advances towards clinical application of HIV-1 GT.

PMID: 21504389

Expert Opin Biol Ther. 2011 Apr 20. [Epub ahead of print]

Recombinant AAV-directed gene therapy for type I glycogen storage diseases.

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Introduction: Glycogen storage disease (GSD) type Ia and Ib are disorders of impaired glucose homeostasis affecting the liver and kidney. GSD-Ib also affects neutrophils. Current dietary therapies cannot prevent long-term complications. In animal studies, recombinant adeno-associated virus (rAAV) vector-mediated gene therapy can correct or minimize multiple aspects of the disorders, offering hope for human gene therapy. Areas covered: A summary of recent progress in rAAV-mediated gene therapy for GSD-I; strategies to improve rAAV-mediated gene delivery, transduction efficiency and immune avoidance; and vector refinements that improve expression. Expert opinion: rAAV-mediated gene delivery to the liver can restore glucose homeostasis in preclinical models of GSD-I, but some long-term complications of the liver and kidney remain. Gene therapy for GSD-Ib is less advanced than for GSD-Ia and only transient correction of myeloid dysfunction has been achieved. A question remains as to whether a single rAAV vector can meet the expression efficiency and tropism required to treat all aspects of GSD-I, or if a multi-pronged approach is needed. An understanding of the strengths and weaknesses of rAAV vectors in the context of strategies to achieve efficient transduction of the liver, kidney and hematopoietic stem cells is required for treating GSD-I.

PMID: 21504271

Acta Otolaryngol. 2011 Apr 19. [Epub ahead of print]

Fas ligand gene transfer effectively induces apoptosis in head and neck cancer cells.

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Abstract Conclusion: Fas ligand (FasL) gene therapy may provide a new efficient therapeutic model for head and neck squamous cell cancer (HNSCC). Acid ceramidase (AC) may not play an important role in the sensitivity of HNSCC cell lines to Fas-induced apoptosis. Objectives: The aims of this study were to investigate the efficacy of FasL gene therapy for HNSCC in vitro and to determine whether the expression of AC in different kinds of HNSCC cell lines is related to the sensitivity of HNSCC cell lines to Fas-mediated apoptotic induction. Methods: Three HNSCC cell lines (Hep-2, MMSI, and SCCVII) were transfected with pEGFP-FasL, a plasmid containing a modified human FasL gene fused to enhanced green fluorescent protein (GFP). pEGFP-C1, a plasmid containing the GFP gene alone, was used as a control. Cell death was observed by fluorescence imaging and quantified using a tetrazolium-based (MTS) assay. SCCVII cells were analyzed by flow cytometry to determine the presence of apoptotic induction. Hep-2 and MMSI cells were evaluated by quantitative real-time PCR to evaluate the expression of AC. Results: Transfection of pEGFP-FasL plasmid was shown to be able to induce cell death, the sensitivity of Fas-mediated apoptosis in HNSCC was different, and the level of AC did not correlate with the sensitivity of HNSCC cells to Fas-induced apoptosis.

PMID: 21502042

PDA J Pharm Sci Technol. 2010 Sep-Oct;64(5):386-91.

Application of lentiviral vectors for development of production cell lines and safety testing of lentiviral-derived cells or products.

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Lentiviral vectors (LVs) are frequently used to engineer cell lines for preclinical research purposes including assay development and target validation. Development of production cell lines for manufacturing recombinant protein therapeutics may also benefit from the use of LVs because they may reduce timelines and generate more uniform or higher expressing stable pools and clones. In addition, LVs could be advantageous for engineering new, alternative host cell substrates due to their ability to efficiently transduce most cell types. We demonstrate here that NS0 mouse myeloma cells, a host cell frequently used for protein production, can be transduced with LVs to greater than 80% efficiency and with no cytotoxic effects. The use of LVs for engineering of production cell lines will require additional testing procedures. Since LVs have previously been used in human gene therapy clinical trials, safety testing assays and procedures have been developed that could easily be applied to the development process for manufacturing cell lines to ensure the absence of unwanted viral material in cell banks and biologic products.

PMID: 21502041

PDA J Pharm Sci Technol. 2010 Sep-Oct;64(5):379-85.

Lentiviral vector-mediated genetic modification of cell substrates for the manufacture of proteins and other biologics.

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Transduction with Lentiviral vectors has been shown to be the most efficient method for the stable delivery of nucleic acid sequences into mammalian cells. Lentiviral vectors have been widely used in research and have recently shown success in clinical trials for human gene therapy. In this paper, we describe the use of lentiviral vectors to generate genetically modified cell substrates for the manufacture of proteins and other complex biologics. The use of lentiviral vectors for the generation of genetically modified cell substrates for the production of biologic material has several advantages over other systems: (1) highly productive mammalian cell lines can be rapidly generated without selection or gene amplification; (2) the high number of vector copies are distributed throughout the open chromatin of the genome, resulting in cell lines that are extremely stable for high levels of gene expression and, consequently, protein production; and (3) high levels of protein glycosylation are maintained despite very high levels of protein production. These advantages offer the potential to significantly improve the quality, time-to-market, and manufacturing cost of biologics for human use.

PMID: 21501423

CNS Neurosci Ther. 2011 Feb 16. doi: 10.1111/j.1755-5949.2011.00246.x. [Epub ahead of print]

Screening of Therapeutic Strategies for Huntington's Disease in YAC128 Transgenic Mice.

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Huntington's disease (HD) is a hereditary neurodegenerative disorder caused by an unstable expansion of cytosine-adenine-guanine (CAG) repeats in the HD gene. The symptoms include cognitive dysfunction and severe motor impairment with loss of voluntary movement coordination that is later replaced by bradykinesia and rigidity. The neuropathology is characterized by neuronal loss mainly in the striatum and cortex, and the appearance of neuronal intranuclear inclusions of mutant huntingtin. The mechanisms responsible for neurodegeneration are still not fully understood although excitotoxicity and a consequent increase in intracellular calcium concentration as well as the activation of caspases and calapins are known to play a key role. There is currently no satisfactory treatment or cure for this disease. The YAC128 transgenic mice express the full-length human HD gene with 128 CAG repeats and constitute a unique model for the study of HD as they replicate the slow and biphasic progression of behavioral deficits characteristic of the human condition and show striatal neuronal loss. As such, these transgenic mice have been an invaluable model not only for the elucidation of the neurodegenerative pathways in HD, but also for the screening and development of new therapeutic approaches. Here, I will review the unique characteristics of this transgenic HD model and will provide a summary of the therapies that have been tested in these mice, namely: potentiation of the protective roles of wild-type huntingtin and mutant huntingtin aggregation, transglutaminase inhibition, inhibition of glutamate- and dopamine-induced toxicity, apoptosis inhibition, use of essential fatty acids, and the novel approach of intrabody gene therapy. The insights obtained from these and future studies will help identify potential candidates for clinical trials and will ultimately contribute to the discovery of a successful treatment for this devastating neurodegenerative disorder.

PMID: 21499927

Med Oncol. 2011 Apr 17. [Epub ahead of print]

Nanoparticles targeting HLA-G for gene therapy in cancer.

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Source
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Cancer cells are aided by immune-tolerant functions of HLA-G to escape the immune surveillance. In general, cancer cells can express membranous HLA-G, secrete soluble HLA-G, produce HLA-G positive exosomes, and can be subjected to proteolytic cleavage by matrix metalloproteinases releasing shedding HLA-G1 in stressful conditions. Thus, the downregulation of HLA-G either in transcripts or proteins may affect positively cancer therapy. The aim of this study was to examine the molecular nanoparticles targeting HLA-G. Special focus was accorded to RNA interference particles. Although numerous studies have reported the importance of HLA-G gene expression modulation by nanoparticles, no studies have investigated clinically their efficiency in this modulation.

PMID: 21499319

Bone Marrow Transplant. 2011 Apr 18. [Epub ahead of print]

Allogeneic cellular gene therapy in hemoglobinopathies-evaluation of hematopoietic SCT in sickle cell anemia.

Lucarelli G, Gaziev J, Isgrò A, Sodani P, Paciaroni K, Alfieri C, De Angelis G, Marziali M, Simone MD, Gallucci C, Roveda A, Saltarelli F, Torelli F, Andreani M.
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Many patients with thalassemia have been cured with BMT since the first successful transplant in 1981. Allogeneic stem cell gene therapy is the only treatment option for patients with sickle cell anemia (SCA). A total of 11 patients with a median age of 12 years (range, 2-16), affected by SCA, received hematopoietic SCT from HLA-identical, related donors following a myeloablative-conditioning regimen. Indications for transplantation were vaso-occlusive crisis, acute chest syndrome, avascular bone necrosis, chronic RBC transfusions, or hemorrhagic stroke. All patients had sustained engraftment. One patient became a stable mixed chimera with 25% of donor cells at 4 years after transplantation. One patient died at 1 year after transplantation. The probability of survival, SCA-free survival and TRM at 5 years after transplant were 90, 90 and 10%, respectively. All 10 surviving patients remained free of any SCA-related events after transplantation. In conclusion, these data confirm SCT from a suitable HLA-matched, related donor should become the primary option for curing children with SCA. There is an excellent survival rate and a return to normal life, free of SCA-related events.

PMID: 21499295

Nat Rev Genet. 2011 May;12(5):341-55.

Therapeutic in vivo gene transfer for genetic disease using AAV: progress and challenges.

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In vivo gene replacement for the treatment of inherited disease is one of the most compelling concepts in modern medicine. Adeno-associated virus (AAV) vectors have been extensively used for this purpose and have shown therapeutic efficacy in a range of animal models. Successful translation to the clinic was initially slow, but long-term expression of donated genes at therapeutic levels has now been achieved in patients with inherited retinal disorders and haemophilia B. Recent exciting results have raised hopes for the treatment of many other diseases. As we discuss here, the prospects and challenges for AAV gene therapy are to a large extent dependent on the target tissue and the specific disease.

PMID: 21499127

J Immunother. 2011 May;34(4):343-52.

Genetic Engineering of Murine CD8+ and CD4+ T Cells for Preclinical Adoptive Immunotherapy Studies.

Kerkar SP, Sanchez-Perez L, Yang S, Borman ZA, Muranski P, Ji Y, Chinnasamy D, Kaiser AD, Hinrichs CS, Klebanoff CA, Scott CD, Gattinoni L, Morgan RA, Rosenberg SA, Restifo NP.
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T-cell receptor (TCR) gene therapy enables for the rapid creation of antigen-specific T cells from mice of any strain and represents a valuable tool for preclinical immunotherapy studies. Here, we describe the superiority of γ-retroviral vectors compared with lentiviral vectors for transduction of murine T cells and surprisingly illustrate robust gene-transfer into phenotypically naive/memory-stem cell like (TN/TSCM; CD62L/CD44) and central memory (TCM; CD62L/CD44) CD8 T cells using murine stem cell-based γ-retroviral vectors (MSGV1). We created MSGV1 vectors for a major histocompatibility complex-class I-restricted TCR specific for the melanocyte-differentiation antigen, glycoprotein 100 (MSGV1-pmel-1), and a major histocompatibility complex-class II-restricted TCR specific for tyrosinase-related protein-1 (MSGV1-TRP-1), and found that robust gene expression required codon optimization of TCR sequences for the pmel-1 TCR. To test for functionality, we adoptively transferred TCR-engineered T cells into mice bearing B16 melanomas and observed delayed growth of established tumors with pmel-1 TCR engineered CD8 T cells and significant tumor regression with TRP-1 TCR transduced CD4 T cells. We simultaneously created lentiviral vectors encoding the pmel-1 TCR, but found that these vectors mediated low TCR expression in murine T cells, but robust gene expression in other murine and human cell lines. These results indicate that preclinical murine models of adoptive immunotherapies are more practical using γ-retroviral rather than lentiviral vectors.