Antioxidant poly(vanillyl alcohol-oxalate) nanoparticles as novel therapeutics for acetaminophen-induced acute liver failure

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ABSTRACT SUMMARY

Acute liver failure (ALF) is the appearance of severe complications rapidly after the first sign of liver disease. ALF has sustained severe damage due to the reactive oxygen species (ROS) including hydrogen peroxide (H₂O₂), which leads to the oxidative stress and the subsequent functional decline of organ systems. We developed novel fully biodegradable poly(vanillyl alcohol-oxalate) (PVAX), in which antioxidant vanillyl alcohol and H₂O-reacting peroxalate ester groups are incorporated in its backbone. The potential of PVAX particles as drug carriers and antioxidant therapeutics was evaluated using a mouse model of acetaminophen (APAP)–induced ALF.

INTRODUCTION

ROS are an essential physiology regulator and serve as an important biological messenger in cell signal transduction cascades. However, overproduction of ROS leads to oxidative stress, resulting in significant damage to cell structures such as lipid membranes, protein and DNA and eventually cause of acute inflammatory diseases. ALF is a complex multi-systemic illness and is more dramatic or more devastating than any other conditions in medicine, responsible for thousands of death each year in world. The common cause of ALF includes viral hepatitis, drug- or toxin-induced liver diseases and metabolic disorder. Among them, APAP-overdose has recently been the most frequent cause of ALF in United State and most European countries. APAP is the most widely used analgesic and its overdose, intentional or unintentional, is known to cause massive hepatocellular apoptosis and hemorrhagic necrosis, leading to ALF.

There has been great interest in the use of antioxidant and anti-inflammatory drugs for the treatment of ALF. However, their therapeutic applications have been curtailed by inconsistent effectiveness resulting from low bioavailability and lack of ability to target macrophages in liver.

We have developed a new family of biodegradable and antioxidant polymers, PVAX, which is able to scavenge H₂O₂ and releases vanillyl alcohol with potent antioxidant and anti-inflammatory activities. Manganese porphyrin (MnP) is one of metalloporphyrins which have emerged as a novel class of catalytic antioxidants that scavenge a wide range of ROS such as superoxide and peroxynitrite. In this work, we developed MnP-loaded PVAX particles and evaluate their potential as antioxidant and anti-inflammatory therapeutics using a mouse model of APAP-induced ALF.

EXPERIMENTAL METHODS

PVAX was synthesized with 1,4-cyclohexane dimethanol, vanillyl alcohol and oxalyl chloride. PVAX particles were prepared by a single emulsion method and MnP-loaded PVAX particles were prepared by a double emulsion method. The morphology of PVAX particles were investigated by scanning electron microscope (SEM). We assessed the H₂O₂ scavenging ability of PVAX particles using Amplex Red assay. The cytotoxicity of PVAX particles were evaluated using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazoliu-m bromide (MTT) assay. Drug release kinetics of MnP-loaded PVAX particles was determined using a UV spectrometer. We investigated the antioxidant and anti-inflammatory activities of MnP-loaded PVAX particles in LPS lipopolysaccharide)-treated macrophages RT-PCR (reverse transcription polymerase chain reaction). A mouse model of APAP-induced ALF was used to evaluate the potential of PVAX particles as therapeutics and drug carriers for MnP in vivo. Mice were treated with various amounts of MnP-loaded PVAX particles 1 h prior to APAP injection. ALF was achieved by intraperitoneal injection of APAP. Mice were sacrificed 23 h after APAP intoxication and whole blood and livers were collected. The activity of ALT in serum was determined using an ALT enzymatic assay kit. Histological sections were made and stained with hematoxylin and eosin (H&E).
RESULTS AND DISCUSSION

Dynamic light scattering revealed that PVAX particles have an average diameter of $\sim 450$ nm. We first investigated the ability of PVAX particles to scavenge $\text{H}_2\text{O}_2$. PVAX particles significantly reduced the $\text{H}_2\text{O}_2$ concentration than free vanillyl alcohol, demonstrating the potential as $\text{H}_2\text{O}_2$ scavengers. The MTT assay reveals that PVAX particles exhibited remarkable biocompatibility profiles. Vanillyl alcohol showed modest anti-inflammatory activities by suppressing the generation of pro-inflammatory cytokines such as $\text{IL}$($\text{interleukin}$)-6, IL-1$\beta$, iNOS (inducible nitric oxide synthase) and COX(cyclooxygenase)-2. However, PVAX particles showed the stronger anti-inflammatory activity than vanillyl alcohol due mainly to their strong antioxidant activity (Fig. 1).

Concentration

CONCLUSION

We evaluated the potential of fully biodegradable and biocompatible PVAX particles as drug delivery systems for treating ALF. The ability of PVAX particles to deliver drug was evaluated using a mouse model of APAP-induced ALF and MnP as a model drug. MnP delivered by PVAX particles significantly reduced the hepatic cellular damages, evidenced by the ALF assay and histological studies. The excellent biocompatibility and rapid drug release profile make PVAX a promising drug delivery system for the treatment of acute inflammatory diseases.

REFERENCES


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