## Abstract

Sex differentiation of the male reproductive tract in mammals is driven, in part, by fetal androgen production. In utero, some phthalate esters (PEs) alter fetal Leydig cell differentiation, reducing the expression of several genes associated with steroid synthesis/transport, and consequently, lowering fetal androgen and Insl3 hormone levels. Simvastatin (SMV) is a cholesterol-lowering drug that directly inhibits HMG-CoA reductase. SMV may also disrupt steroid biosynthesis, but through a different mode of action (MOA) than the PEs. As cholesterol is a precursor of steroid hormone biosynthesis, we hypothesized that in utero exposure to SMV during the critical period of sex differentiation would lower fetal testicular testosterone (T) production without affecting genes involved in cholesterol and androgen synthesis and transport. Secondly, we hypothesized that a mixture of SMV and a PE, which may have different MOAs, would reduce testosterone levels in an additive manner. Pregnant Sprague Dawley rats were dosed orally with SMV, dipentyl phthalate (DPeP), or SMV plus DPeP from gestational days 14-18, and fetuses were evaluated on GD18. On GD18, SMV lowered fetal T production and serum triglycerides, low density lipoprotein, high density lipoprotein, and total cholesterol levels, and downregulated two genes in the fetal testis that were different from those altered by PEs. When SMV and DPeP were administered as a mixture, fetal T production was significantly reduced in an additive manner, thus demonstrating that a mixture of chemicals can induce additive effects on fetal T production even though they display different MOAs.