BACKGROUND
Selenium is an essential trace element that is incorporated as selenocysteine into the primary structure of selenoproteins. Nutritional deficiency of selenium decreases selenoprotein concentrations and leads to pathologic conditions. Most of the known selenoproteins are members of the glutathione peroxidase or iodothyronine deiodinase families. Selenoprotein P (SEPP1) is a major selenoprotein that is not a member of those families. It is an extracellular glycoprotein that is present in several isoforms and is the only selenoprotein known to contain multiple selenocysteine residues. It is a heparin-binding protein that appears to be associated with endothelial cells and has been implicated as an oxidant defense in the extracellular space. Although there is evidence of several isoforms of the protein, all of them share the same amino-terminal sequence and therefore are likely products of the same gene. The gene which encodes Selenoprotein P maps to human chromosome 5q31.

REFERENCES

CHROMOSOMAL LOCATION
Genetic locus: SEPP1 (human) mapping to 5q31.

PRODUCT
Selenoprotein P siRNA (h) is a pool of 3 target-specific 20-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections.

For independent verification of Selenoprotein P (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-40930A, sc-40930B and sc-40930C.

STORAGE AND RESUSPENSION
Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNase-free water provided. Resuspension of the siRNA duplex in 330 µl of RNase-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS
Selenoprotein P siRNA (h) is recommended for the inhibition of Selenoprotein expression in human cells.

Selenoprotein P (C-14): sc-22639 is recommended as a control antibody for Western Blotting (starting dilution 1:100, dilution range 1:100-1:1,000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) protein detection using the recommended secondary reagents listed below.

RECOMMENDED SECONDARY REAGENTS
To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2783 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

SUPPORT REAGENTS
For optimal siRNA transfection efficiency, Santa Cruz Biotechnology’s siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10µM in 60 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238. Semi-quantitative RT-PCR may be performed using RT-PCR Primer: Selenoprotein P (h)-PR: sc-40930-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE
For research use only, not for use in diagnostic procedures.

PROTOCOLS
See our web site at www.scbt.com or our catalog for detailed protocols and support products.