and Related Metab. Dis., 19, 458-463 (1995)). However, at elevated dosage levels, beneficial effects are reversed and dangerous toxicity is manifested.

Research over the last two decades has suggested that selenium is effective in the reduction of cancer incidence when provided to animals at doses only 5- to 10-fold above nutritional requirement (See, e.g., El-Bayoumy, The role of selenium in cancer prevention, Philadelphia, Lippincott, 1-15, 1991). Chemoprevention studies with selenium in animal model systems have indicated that this element is effective for most, if not all of the organ systems and is protective against carcinogenic effects (See, e.g., El-Bayoumy, The role of selenium in cancer prevention, Philadelphia, Lippincott, 1-15, 1991. Both epidemiological studies and supplementation trials have also supported its efficacy in lowering the incidence of cancers of the liver, colon, prostate and lung (See, e.g., Yu et al. Biol Trace Elem Res, 56: 117-124 (1997); Clark et al., J Am Med Assoc, 276: 1957-1963 (1996); Yoshizawa et al., J Natl Cancer Inst, 90: 1219-1224, (1998); Brooks, et al., J Urol, 166: 2034-2038, (2001)). Other studies have demonstrated no beneficial effect for selenium reduction of cancers (See, e.g., Garland et al., J. Am. Coll Nutr., 12: 400-11 (1993); Ghadirian et al., Cancer Detect Prev, 24: 305-13 (2000)).

Multiple forms of selenium have been examined. These include inorganic selenium such as sodium selenite, and organic sources, including selenium yeast. There is a significant difference between toxicity of inorganic and organic selenium, the inorganic compounds usually being absorbed and utilized less efficiently and also being more toxic than organic sources of selenium.

SUMMARY ~~OF THE INVENTION~~

The present ~~invention~~disclosure relates to soluble selenium compositions and methods of production, separation and purification thereof. In particular the present ~~invention~~disclosure provides methods of preparing water soluble selenoglycoproteins (e.g., via extracting selenoglycoproteins from selenium enriched yeast), methods of supplementing a selenium deficient composition via admixing water soluble selenoglycoproteins with the selenium deficient composition, compositions comprising the water soluble selenoglycoproteins and methods of administering the same.

Accordingly, in ~~some embodiments~~an aspect, the present ~~invention~~disclosure provides a method for preparation of soluble selenoglycoproteins comprising: providing selenium enriched yeast ,

exposing the selenium enriched yeast to acidic conditions followed by centrifugation to generate i) a pellet comprising acid insoluble material; and ii) a liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions; precipitating selenoglycoproteins from the liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions via raising the pH of the liquid phase, and (d) separating the precipitated selenoglycoproteins from the liquid phase. In some embodiments, exposing the selenium enriched yeast to acidic conditions occurs at a temperature greater than room temperature (e.g., greater than about 20-25°C). The present ~~invention~~disclosure is not limited by the temperature greater than room temperature under which the selenium enriched yeast are exposed to acidic conditions. Indeed, a variety of temperatures may be used including, but not limited to, about 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 90, 95, 97, 99°C, or hotter). Similarly, the ~~invention~~present disclosure is not limited by the acidic conditions under which the selenium enriched yeast are exposed. In some embodiments, the selenium enriched yeast are exposed to conditions wherein the pH is about 6.5, about 6, about 5.5, about 5, about 4.5, about 4, about 3.5, about 3, about 2.5, about 2, about 1.5, about 1, or less. In a preferred embodiment, exposing the selenium enriched yeast to acidic conditions comprises exposure of the selenium enriched yeast to a pH of 1.5. In some embodiments, exposing the selenium enriched yeast to acidic conditions takes place for a certain period of time. The ~~invention~~present disclosure is not limited by the amount of time under which the selenium enriched yeast are exposed to acidic conditions. In some embodiments, the selenium enriched yeast are exposed to acidic conditions for about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14, 16, 18, 20, 22, 24 or more hours. In some embodiments, the selenium enriched yeast are exposed to acidic conditions for between one and twenty-four hours. In some embodiments, the selenium enriched yeast are exposed to acidic conditions for between five and ten hours. In a preferred embodiment, the selenium enriched yeast are exposed to acidic conditions for about eight hours. In some embodiments, acidic conditions are created and/or maintained utilizing acidic buffer and/or the addition of an acid. The ~~invention~~present disclosure is not limited by the type of acid utilized. Indeed, any acid may be used. In some embodiments, the acid is hydrochloric acid, although any acid may be used . In some embodiments, selenoglycoproteins are precipitated from the liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions via raising the pH of the liquid phase. In some embodiments, the precipitated selenoglycoproteins are separated from the liquid phase via centrifugation to form a pellet of the precipitated

selenoglycoproteins followed by removal of the liquid phase from the precipitated selenoglycoproteins. The ~~invention~~present disclosure is not limited by the amount or degree to which the pH of the extract is raised in order to precipitate the selenoglycoproteins from the liquid phase. In some embodiments, the pH of the liquid phase is raised to 1.85. In some embodiments, the pH of the liquid phase is raised to 3.0. In some embodiments, the pH of the liquid phase is raised to 4.0. In some embodiments, the pH of the liquid phase is raised to 6.0. In some embodiments, selenoglycoproteins are precipitated and separated from the liquid phase at a variety of pH conditions to generate multiple pH dependent fractions of soluble selenoglycoproteins. For example, in some embodiments, a single liquid phase comprising the extract of selenium enriched yeast soluble under acidic conditions is used to create a first fraction of soluble selenoglycoproteins precipitated at a first pH (e.g., pH of 1.85), a second fraction of soluble selenoglycoproteins precipitated at a second pH (e.g., pH of 3.0), a third fraction of soluble selenoglycoproteins precipitated at a third pH (e.g., pH of 4.0), and a fourth fraction of soluble selenoglycoproteins precipitated at a fourth pH (e.g., pH of 6.0). In some embodiments, precipitating the selenoglycoproteins from the liquid phase via raising the pH of the liquid phase comprises multiple, sequential pH dependent precipitation reactions of the liquid phase. In some embodiments, a composition comprising soluble selenoglycoproteins contains only a single, pH dependent fraction of the selenoglycoproteins (e.g., soluble selenoglycoproteins from liquid phase comprising the extract of selenium enriched yeast soluble under acidic conditions precipitated at pH 4.0). In some embodiments, a composition comprising soluble selenoglycoproteins contains two or more pH dependent fractions of selenoglycoproteins (e.g., soluble selenoglycoproteins from liquid phase comprising the extract of selenium enriched yeast soluble under acidic conditions precipitated at two or more different pH values).

In some embodiments, the selenium enriched yeast are dried, nonviable selenium-enriched yeast containing 2% or less inorganic selenium .

The ~~invention~~present disclosure also provides compositions comprising soluble selenoglycoproteins prepared according to the ~~invention~~present disclosure. For example, in some embodiments, the ~~invention~~present disclosure provides a selenium deficient composition comprising soluble selenoglycoproteins of the ~~invention~~present disclosure. In some embodiments, soluble selenoglycoproteins of the ~~invention~~present disclosure are added to and/or mixed with a selenium deficient composition. The ~~invention~~present disclosure is not limited by the type of selenium deficient composition or material to which soluble selenoglycoproteins of the ~~invention~~present disclosure are added to or

mixed with, such compositions or materials including, but not limited to, dietary supplements, drugs, pharmaceuticals, foodstuffs, animal feeds, and other types of materials. In some embodiments, adding or mixing (e.g., admixing) selenoglycoprotein comprises admixing selenoglycoprotein and one or more other types of selenium into the composition. In some embodiments, soluble selenoglycoproteins of the ~~invention~~present disclosure are added to and/or mixed with compositions that contain selenium (e.g., to augment and/or supplement the total amount of selenium).

The ~~invention~~present disclosure also provides a composition comprising soluble selenoglycoproteins and a carrier. In some embodiments, the selenoglycoproteins are a specific pH dependent fraction of selenoglycoproteins. The ~~invention~~present disclosure is not limited by the type of carrier utilized. Indeed a variety of carriers may be used including, but not limited to, dendrimers, polymerosomes, nanoparticles, slow-release polymers, nanocapsules, and/or molecularly imprinted polymers. In one preferred embodiment, the carrier is a slow-release polymer. In another preferred embodiment, the carrier is a molecularly imprinted polymer. In still another preferred embodiment, the carrier is a polymersome used to encapsulate selenoglycoprotein. The ~~invention~~present disclosure is not limited by the type of polymersome used. Indeed, any polymersome known in the art may be utilized. In some embodiments, the polymersome comprises poly(ethylene oxide) (PEO) block copolymer. However, the ~~invention~~present disclosure is not so limited. Any known block copolymer may be utilized, including, for example poly(ethylethylene) (PEE), poly(butadiene) (PB or PBD), poly(styrene) (PS), and poly(isoprene) (PI). In some embodiments, the polymer comprises poly(ε-caprolactone) (PCL) diblock co-polymer. In some embodiments, the polymersome comprises poly(ethylene oxide)-block-poly(ε-caprolactone) (PEO-b-PCL) based diblock copolymers. In some embodiments, the polymersome comprises a block copolymer that is a triblock, tetrablock, pentablock, or at least six block copolymer. In some embodiments, the polymersome is derived from the coupling of poly(lactic acid), poly(glycolide), poly(lactic-coglycolic acid) and/or or poly(3-hydroxybutyrate) with PEO. The ~~invention~~present disclosure is not limited by the size of a polymersome encapsulated selenoglycoprotein. A variety of sizes find use in the compositions and methods of the ~~invention~~present disclosure including, but not limited to, polymersome encapsulated selenoglycoproteins that are about, 50-300 nm in diameter although larger (e.g., about 350 nm, 400 nm, 500 nm or larger) and smaller (e.g., about 40 nm, 30 nm. 20 nm, or smaller) polymersome encapsulated selenoglycoproteins may be used.

CLAIMS

What Is Claimed Is:

1. A composition comprising soluble selenoglycoproteins, wherein the soluble selenoglycoproteins are obtained via acid extraction of the soluble selenoglycoproteins from selenium enriched yeast.

2. The composition of claim 1, wherein the soluble selenoglycoproteins are obtained, subsequent to exposing the selenium enriched yeast to acidic conditions, via pH dependent precipitation of the soluble selenoglycoproteins.

3. The composition of claim 2, wherein pH dependent precipitation comprises raising the pH to a level at which a portion of the soluble selenoglycoproteins precipitate out of a liquid phase comprising the extract of selenium enriched yeast soluble under acidic conditions and collecting the precipitated selenoglycoproteins.

4. The composition of claim 3, wherein the pH level is raised to 4.0.

5. The composition of claim 3, wherein the pH level is raised to 6.0.

6. The composition of claim 1, wherein the soluble selenoglycoproteins are generated by a method comprising:

a) providing selenium enriched yeast;

b) exposing the selenium enriched yeast to acidic conditions followed by centrifugation to generate

i) a pellet comprising acid insoluble material; and

ii) a liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions;

c) precipitating selenoglycoproteins from the liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions via raising the pH of the liquid phase; and

d) separating the precipitated selenoglycoproteins from the liquid phase.

7. A composition comprising soluble selenoglycoproteins and a carrier.

8. The composition of claim 7, wherein the carrier is selected from the group consisting of a polymersome, a slow-release polymer, a nanocapsule, and a molecularly imprinted polymer.

9. The composition of claim 7, wherein the carrier is a slow-release polymer.

10. The composition of claim 7, wherein the carrier is a polymersome used to encapsulate selenoglycoprotein.

11. The composition of claim 10, wherein the polymersome comprises poly(ethylene oxide) (PEO) block copolymer.

12. The composition of claim 10, wherein the polymersome comprises poly(ε-caprolactone) (PCL) diblock copolymer.

13. The composition of claim 10, wherein the polymersome comprises poly(ethylene oxide)-block-poly (ε-caprolactone) (PEO-b-PCL) based diblock copolymers.

14. The composition of claim 10, wherein the polymersome is derived from the coupling of poly(lactic acid), poly(glycolide), poly(lactic-coglycolic acid) or poly(3-hydroxybutyrate) with PEO.

15. The composition of claim 10, wherein the average diameter of a polymersome encapsulating selenoglycoprotein is 50-300 nm.

16. A method for preparation of soluble selenoglycoproteins comprising:

a) providing selenium enriched yeast;

b) exposing the selenium enriched yeast to acidic conditions followed by centrifugation to generate

i) a pellet comprising acid insoluble material; and

ii) a liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions;

c) precipitating selenoglycoproteins from the liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions via raising the pH level of the liquid phase; and

d) separating the precipitated selenoglycoproteins from the liquid phase.

17. The method of claim 16, wherein the acidic conditions are obtained using acidic buffer or the addition of an acid.

18. The method of claim 17, wherein the acid is hydrochloric acid.

19. The method of claim 16, wherein exposing the selenium enriched yeast to acidic conditions comprises exposing the selenium enriched yeast to a pH level of 1.5.

20. The method of claim 16, wherein the selenium enriched yeast are exposed to acidic conditions for between one and twenty-four hours.

21. The method of claim 16, wherein the selenium enriched yeast are exposed to acidic conditions for about 8 hours.

22. The method of claim 16, wherein exposing the selenium enriched yeast to acidic conditions occurs at a temperature greater than room temperature.

23. The method of claim 22, wherein the temperature is between 50 °C and 100 °C.

24. The method of claim 22, wherein the temperature is 80 °C.

25. The method of claim 16, wherein selenoglycoproteins are precipitated from the liquid phase at a variety of pH conditions to generate multiple pH dependent fractions of soluble selenoglycoproteins.

26. The method of claim 25, wherein the multiple pH dependent fractions of soluble selenoglycoproteins are generated at pH values of 1.85, 3.0, 4.0 and 6.0.

27. The method of claim 16, wherein separating the precipitated selenoglycoproteins from the liquid phase comprises centrifugation to form a pellet of the precipitated selenoglycoproteins followed by removal of the liquid phase from the precipitated selenoglycoproteins.

28. The method of claim 16, wherein the selenium enriched yeast are dried, nonviable selenium-enriched yeast containing 2% or less inorganic selenium.

CLAIMS

1. A method for preparation of soluble selenoglycoproteins comprising:

a) providing selenium enriched yeast;

b) exposing the selenium enriched yeast to acidic conditions followed by centrifugation to generate

i) a pellet comprising acid insoluble material; and

ii) a liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions;

c) precipitating selenoglycoproteins from the liquid phase comprising the extract of selenium enriched yeast soluble under the acidic conditions via raising the pH of the liquid phase; and

d) separating the precipitated selenoglycoproteins from the liquid phase.

2. The method of claim 1, wherein the acidic conditions are obtained using acidic buffer or the addition of an acid.

3. The method of claim 2, wherein the acid is hydrochloric acid.

4. The method of claim 1, wherein exposing the selenium enriched yeast to acidic conditions comprises exposing the selenium enriched yeast to a pH between 1.5 and pH 6.5.

5. The method of claim 4, wherein the pH is 1.5.

6. The method of claim 1, wherein the selenium enriched yeast are exposed to acidic conditions for between one and twenty-four hours.

7. The method of claim 1, wherein the selenium enriched yeast are exposed to acidic conditions for between five and ten hours.

8. The method of claim 1, wherein the selenium enriched yeast are exposed to acidic conditions for about 8 hours.

9. The method of claim 1, wherein exposing the selenium enriched yeast to acidic conditions occurs at a temperature greater than room temperature.

10. The method of claim 9, wherein the temperature is between 50° C. and 100° C.

11. The method of claim 9, wherein the temperature is between 70° C. and 85° C.

12. The method of claim 9, wherein the temperature is 80° C.

13. The method of claim 1, wherein selenoglycoproteins are precipitated from the liquid phase at a variety of pH conditions to generate multiple pH dependent fractions of soluble selenoglycoproteins.

14. The method of claim 13, wherein the multiple pH dependent fractions of soluble selenoglycoproteins are generated at pH values of 1.85, 3.0, 4.0 and 6.0.

15. The method of claim 1, wherein separating the precipitated selenoglycoproteins from the liquid phase comprises centrifugation to form a pellet of the precipitated selenoglycoproteins followed by removal of the liquid phase from the precipitated selenoglycoproteins.

16. The method of claim 1, wherein the method is used to create a fraction of soluble selenoglycoproteins precipitated at a value selected from the group consisting of pH of 1.85, 3.0, 4.0 and 6.0.

17. The method of claim 1, wherein the selenium enriched yeast are dried, nonviable selenium-enriched yeast containing 2% or less inorganic selenium.