

WHAT IS CLAIMED IS:

1. ~~An egg yolk extract comprising free yolk granules.~~
2. ~~An egg yolk extract comprising the liquid contents of an egg yolk and one or more optionally lysed yolk granules.~~
3. ~~An egg yolk extract comprising the liquid contents of yolk spheres.~~
4. ~~An egg yolk extract comprising the liquid contents of yolk granules.~~
5. ~~An egg yolk extract comprising the liquid contents of an egg yolk and one or more egg white proteins.~~
6. ~~The egg yolk extract of claim 5, wherein the one or more egg white proteins comprise ovalbumin, ovotransferrin, lysozyme, ovalbumin-related protein X (OVAX), defensins, ovoinhibitor, AvBD11, or a combination thereof.~~
71. An egg yolk extract that supports cell proliferation and/or survival at least equivalent to FBS when used at the same concentration in a cell culture medium.
82. The egg yolk extract of ~~any one of claims 2 to 71~~, comprising free yolk granules and/or
9. ~~The egg yolk extract of any one of claims 1 to 8~~, comprising lysed yolk spheres.
103. The egg yolk extract of ~~any one of claims 7 to 9~~ 1 or 2, comprising one or more egg white proteins such as ovalbumin, ovotransferrin, lysozyme, ovalbumin-related protein X (OVAX), defensins, ovoinhibitor, AvBD11, or a combination thereof.
114. The egg yolk extract of any one of claims 1 to ~~103~~, comprising the homogenized yolk and white of an egg.
125. The egg yolk extract of any one of claims 1 to ~~114~~, wherein the egg yolk extract consists essentially of egg yolk and egg white, wherein the egg yolk comprises lysed yolk spheres; preferably.
13. ~~The egg yolk extract of any one of claims 1 to 12~~, wherein the egg yolk extract is liquid.
146. The egg yolk extract of any one of claims 1 to ~~135~~, wherein the egg yolk extract is extracted from an egg of any oviparous species, for example, birds, reptiles, amphibians, fish, insects, molluscs, arachnids, or combinations thereof; or.

~~15. The egg yolk extract of any one of claims 1 to 13, wherein the egg yolk extract is avian, or-~~

~~16. The egg yolk extract of claim 15, wherein the egg yolk extract is extracted from an egg of a chicken, turkey, duck, goose, quail, pheasant, ostrich, emu, or combinations thereof, or -~~

~~17. The egg yolk extract of claim 16, wherein the egg yolk extract is chicken, duck, or quail.~~

~~187. A supplement for cell culture medium, the supplement comprising the egg yolk extract of any one of claims 1 to 176.~~

~~198. The supplement of claim 187, consisting essentially of the egg yolk extract of any one of claims 1 to 176.~~

~~209. The supplement of claim 198, consisting of the egg yolk extract of any one of claims 1 to 166.~~

~~21. The supplement of any one of claims 1 to 20, wherein the yolk extract is not diluted.~~

~~22. The supplement of claim 21, wherein the yolk extract is not diluted with saline, such as PBS.~~

~~2310. A cell culture medium comprising the egg yolk extract of any one of claims 1 to 166 or the supplement of any one of claims 187 to 229.~~

~~24. The cell culture medium of claim 23, wherein the medium is Dulbecco's Modified Eagle Medium (DMEM), RPMI media, CMRL1066, Hanks' Balanced Salt Solution (HBSS) phosphate buffered saline (PBS), L-15 medium, DMEM-F12, EpiLife® medium, and Medium 171, or a combination thereof.~~

~~25. The cell culture medium of claim 24, wherein the medium is a buffered saline solution, such as HBSS.~~

~~2611. The cell culture medium of any one of claims 23 to 2510, comprising from about 1% to about 10% v/v of the extract or supplement, such as about 1%, about 2%, about 3%, about 4%, about 5%, about 6%, about 7%, about 8%, about 9%, or about 10%.~~

~~2712. The cell culture medium of any one of claims 23 to 2610 or 11, wherein the cell culture medium does not contain FBS and/or NCS.~~

~~2813.~~ The cell culture medium of any one of claims ~~23-10~~ to ~~2712~~, wherein the cell culture medium is a growth medium or a freezing medium, for example a freezing medium without DMSO.

~~2914.~~ The cell culture medium of any one of claims ~~23-10~~ to ~~2813~~, wherein the cell culture medium is for culturing primary cell lines, for example, primary cell lines for food consumption, for example, primary bovine skeletal muscle cells, or:

~~30.~~ ~~The cell culture medium of any one of claims 23 to 29, wherein the cell culture medium is for use in culturing cells for antibody production-,~~ or

~~31.~~ ~~The cell culture medium of any one of claims 23 to 29, wherein the cell culture medium is for use in biologics production, for example, biologics such as insulin, erythropoietin (EPO), or Granulocyte Colony Stimulating Factor (C-GSF).~~

~~3215.~~ A method for making an egg yolk extract, the method comprising lysing yolk spheres.

~~3316.~~ The method of claim ~~3215~~, wherein lysing yolk spheres comprises sonicating or homogenizing the yolk spheres, for example by high pressure homogenization.

~~3417.~~ The method of claim ~~3316~~, wherein the method is sufficient to lyse at least about 50%, about 60%, about 70%, about 80%, about 90%, about 95%, or about 99% of the yolk spheres.

~~3518.~~ The method of claim ~~33-16~~ or ~~3417~~, wherein the sonicating is at about 75W or more for from about 25 to about 30 cycles at about 15 to about 20 seconds per cycle.

~~3619.~~ The method of any one of claims ~~32-15~~ to ~~3518~~, wherein the yolk spheres are in a composition comprising egg yolk and egg white.

~~3720.~~ The method of claim ~~3619~~, further comprising mixing the egg yolk and egg white before lysing the yolk spheres, optionally wherein the mixing is for from about 30 minutes to about 24 hours, at from about 4°C to about 25°C., such as for about 30 minutes at room temperature or for about 12 to 18 hours at 4°C, and optionally wherein the mixing is by inversion.

~~3821.~~ The method of claim ~~36-19~~ or ~~3720~~, further comprising removing the vitelline membrane from the egg yolk before or after mixing the egg yolk and egg white.

~~3922.~~ The method of any one of claims ~~32-15~~ to ~~3821~~, further comprising dissolving unlysed yolk spheres and/or yolk granules for example by alkalinizing and/or salinating the lysed yolk spheres and:

~~40. The method of claim 39,~~ wherein alkalinizing and/or salinating the lysed yolk spheres comprises adding a base and/or salt to the lysed yolk spheres, such as a strong base, such as NaOH (optionally at from about 100 mM to about 1 M), KOH (optionally at from about 100 mM to about 1 M), Na₂SO₄ (optionally at from about 0.05 mM to about 0.1 M), (NH₄)₂SO₄ (optionally at from about 0.05 mM to about 0.5 M), or a combination thereof or such as NaCl, optionally at from about 0.1 M to about 0.5M.

~~41~~23. The method of any one of claims ~~32-15~~ to ~~40~~22, further comprising removing solids-
and

~~42. The method of claim 41,~~ wherein removing solids comprises centrifuging the lysed yolk spheres, for example at from about 15,000 g to about 30,000 g, and retaining the supernatant.

~~43~~24. The method of any one of claims ~~32-15~~ to ~~42~~23, further comprising sterilizing the egg yolk extract, for example, using a filter with a pore size of 0.4 µm or smaller, such as about a 0.22 µm or 0.11 µm pore size and/or -

~~44. The method of any one of claims 32 to 43,~~ further comprising clarifying the egg yolk extract, for example by adding glycerol at from about 50 to about 200 µL per 1 mL of egg yolk extract.

~~45~~25. An egg yolk extract made by the method of any one of claims ~~32-15~~ to ~~44~~24.