has been shown to require various proteins, including, but not limited to, members of the family of bone morphogenetic proteins (BMPs) and members of the TGF-β superfamily of growth factors. Manufacture, isolation, and purification of such proteins is necessarily more complex and expensive than that of small-molecule chemical compounds because proteins are larger, more complex, and are synthesized by a living cell

Thus, there remains a need for small-molecule chemical compounds — especially ones known to be non-toxic — capable of inducing proliferation of progenitor cells into functional bone, cartilage, tendon, and/or ligamentous tissue.

The solution to this technical problem is provided by the embodiments characterized in the claims

BRIEF SUMMARY

~~The present disclosure provides, in one embodiment,~~In one aspect, there is provided an isolated cell in a culture medium, said culture medium comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine. In a further embodiment, the isolated cell is a mammalian cell. In a further embodiment, the isolated cell is a stem cell. In a further embodiment, the stem cell is a mesenchymal stem cell. In a further embodiment, the theobromine, salt or double salt of theobromine, or co-crystal comprising theobromine is present in an amount from 1 to 300 μM.

~~The present disclosure provides, in another embodiment,~~In one aspect, there is provided a method of culturing a cell, the method comprising: a) culturing said cell in a culture medium, wherein said culture medium comprises theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine. In a further embodiment, the cell is a mammalian cell. In a further embodiment, the cell is a stem cell. In a further embodiment, the stem cell is a mesenchymal stem cell. In a further embodiment, the theobromine is from 1 to 300 μM.

~~The present disclosure provides, in another embodiment,~~In one aspect, there is provided a culture medium comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine. In a further embodiment, the theobromine, salt or double salt of theobromine, or co-crystal comprising theobromine is present in an amount from 1 to 300 μM. In a further embodiment, the theobromine, salt or double salt of theobromine, or co-crystal comprising theobromine is from 1 to 100 μM.

~~The present disclosure provides, in another embodiment,~~In one aspect, there is provided the use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine in the manufacture of a medicament for treating osteoporosis, ~~for~~ enhancing bone formation, increasing bone density, increasing interconnections of internal bone, increasing bone

mass, treating cartilage and/or bone defects, increasing fetal birth weight, or combinations thereof, in a subject in need thereof.

~~The present disclosure provides, in another embodiment,~~ In one aspect, there is provided the use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine in the manufacture of a medicament for preventing tooth decay, remineralizing a tooth surface, treating dentine hypersensitivity, or combinations thereof, in a subject in need thereof.

In one aspect, there is provided the use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine in the manufacture of a medicament for improving maternal and/ or child health during the stages of preconception, pregnancy, lactation, and/ or postpartum.

The present disclosure provides, *in* another embodiment, a method for improving maternal and/or child health during the stages of preconception, pregnancy, lactation, and/or postpartum comprising: administering a composition a) to a mother during pregnancy and/or lactation; or b) to a child during the stage of postpartum; wherein the composition comprises theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine. In a further embodiment, the composition benefits at least one of the mother and child, wherein said benefit is selected from the group consisting of increased fetal weight gain, enhanced bone formation, increased bone density, increased interconnections of internal bone, increased bone mass, and combinations thereof.

In one aspect, there is provided the use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine in the manufacture of a medicament for increasing fetal weight gain, enhancing fetal and/ or maternal bone formation, increasing fetal and/ or maternal bone density, increasing interconnections of internal fetal and/ or maternal bone, increasing fetal and/ or maternal bone mass, treating maternal osteoporosis, and combinations thereof.

~~The present disclosure provides, in another embodiment,~~ In one aspect, there is provided a composition comprising: a) demineralized osteogenic bone powder with a particle size of about 250 μrn to about 5 mm; and b) theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine.

In a further embodiment, the present disclosure provides use of a composition comprising: a) demineralized osteogenic bone powder with a particle size of about 250 μm to about 5 mm; and b) theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine.

In one aspect, there is provided the use of the composition as defined herein in the manufacture of a medicament for application to a bone site to promote new bone growth at the site.

In a further embodiment, the present disclosure provides use of a composition comprising a bone cement and theobromine. For example, without intending to be limited, the bone cement may be formed by combining calcium and phosphate precursors in an aqueous solution, which initially forms a paste but then hardens into a hydroxyapatite bone cement when used. The bone cement may be a composition comprising at least one calcium phosphate, at least one sodium phosphate, and theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine.

BRIEF DESCRIPTION OF THE DRAWINGS

For a further understanding of the nature, objects, and advantages of the present disclosure, reference should be had to the following detailed description, read in conjunction with the following drawings, wherein like reference numerals denote like elements.

14. Use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine ~~for the preparation of a pharmaceutical composition~~ in the manufacture of a medicament for treating osteoporosis, enhancing bone formation, increasing bone density, increasing interconnections of internal bone, increasing bone mass, treating cartilage and/ or bone defects, increasing fetal birth weight, or combinations thereof, in a subject in need thereof.

15. Use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine ~~for the preparation of a pharmaceutical composition~~ in the manufacture of a medicament for preventing tooth decay, remineralizing a tooth surface, treating dentine hypersensitivity, or combinations thereof, in a subject in need thereof.

16. Use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine ~~for the preparation of a pharmaceutical composition~~in the manufacture of a medicament for improving maternal and/ or child health during the stages of preconception, pregnancy, lactation, and/ or postpartum.

17. Use of a composition comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine ~~for the preparation of a pharmaceutical composition~~in the manufacture of a medicament for increasing fetal weight gain, enhancing fetal and/ or maternal bone formation, increasing fetal and/ or maternal bone density, increasing interconnections of internal fetal and/ or maternal bone, increasing fetal and/ or maternal bone mass, treating maternal osteoporosis, and combinations thereof.

18. A composition comprising:

a) demineralized osteogenic bone powder with a particle size of about 250 μηι to about 5 mm; and

b) theobromine, a salt or double salt of theobromine, or a co-crystal comprising theobromine.

19. Use of the composition of claim ~~19~~18 ~~for the preparation of a pharmaceutical composition~~in the manufacture of a medicament for application to a bone site to promote new bone growth at the site.