minimally aqueously soluble, and rapidly catabolized, which are not desirable traits for use in insulin compositions. Another agent stated to accelerate insulin absorption is the protease inhibitor aprotinin, but aprotinin is unsuitable for repeated administration due to its potential to induce immunological reactions. D.R. Owens, *et al.*, 25 BR. J. CLIN. PHARMAC. 453-456 (1988). More recently, it has been stated that a faster onset of action compared with existing insulin therapies can be achieved by adding a nicotinic compound, such as nicotinamide, and the amino acid arginine, and optionally glutamic acid. See U.S. Patent No. 8,324,157.

Nevertheless, there remains a need for insulin compositions, intended for use at meal-time, that: have more rapid uptake of insulin from the injection site and more rapid onsets of action than existing insulin products; are stable chemically and physically during prolonged storage to minimize safety risks and to assure dose accuracy and efficacy; do not result in unacceptable levels of injection site pain; and/or require a minimal number of ingredients and/or excipients to achieve improvements in time action while maintaining sufficient stability.

The present invention seeks to provide formulations which meet these needs. In particular, the present invention provides formulations of insulin and treprostinil which have more rapid uptake of insulin into the blood. The present invention also provides a more rapid onset of action and/or absorption than existing commercial insulin products. In addition, the formulations of the present invention are sufficiently stable. In addition, the administration of the formulations of the present invention does not result in unacceptable injection site pain. The formulations of the present invention achieve these effects while using a minimal number of additional components.

The ~~According to a first aspect of the~~ present disclosure ~~invention, there is~~ provide~~d~~ a composition comprising an insulin and treprostinil.

In one aspect of the present disclosure, there is provided a pharmaceutical composition comprising an insulin and treprostinil, wherein the treprostinil concentration is from about 0.01 to about 30 μM.

In another aspect of the present disclosure, there is provided apharmaceutical composition comprising:

a. insulin lispro, in a concentration from about 40 to about 500 IU/mL,

b. treprostinil, in a concentration from about 0.01 to about 30 μM,

c. meta-cresol, in a concentration from about 2.5 to about 3.8 mg/mL,

d. glycerol, in a concentration from about 5 to about 20 mg/mL,

e. zinc, in a concentration from about 0.00525 to about 0.131 mg/mL, and

f. the pH of the composition is from about 7.0 to about 7.8.

In a further aspect of the present disclosure, there is provided a use of a pharmaceutical composition disclosed herein in the manufacture of a medicament for the treatment of diabetes.

In another aspect of the present disclosure, there is provided a pharmaceutical composition disclosed herein for use in therapy.

In a further aspect of the present disclosure, there is provided a pharmaceutical composition disclosed herein for use in the treatment of diabetes.

In another aspect of the present disclosure, there is provided an article of manufacture comprising any one of the pharmaceutical compositions disclosed herein.

In an embodiment, the insulin is selected from the group consisting of human insulin, or a rapid-acting structural variant, mutant, or analog of human insulin, such as insulin lispro, insulin aspart and insulin glulisine. In a preferred embodiment, the insulin is insulin lispro. In an embodiment, the insulin lispro concentration is from about 40 to about 500 IU/mL. Preferably, the insulin lispro concentration is either 100 IU/mL or 200 IU/mL.

The amount of treprostinil in the composition must be sufficient to result in an onset of action and/or absorption that is faster than that of the insulin formulation with no treprostinil. The concentration of treprostinil used in the compositions of the present ~~invention~~ disclosure ranges from about 0.01 to about 30 μM, particularly from about 0.05 to about 26 μM, about 0.05 to 0.3 μM, about 0.1 to 0.5 μM, about 0.3 to 1.5 μM, about 0.5 to 5.1 μM, about 1.5 to 5.9 μM, about 5.1 to 12.8 μM, about 5.9 to 20 μM, about 12.8 to 24 μM, about 20 to 26 μM, or about 26 to 30 μM, particularly either about 0.05, 0.1, 0.3, 0.5, 1.5, 5.1, 5.9, 12.8, 20, 24 or 26 μM. The insulin and treprostinil are present in a fixed dose combination.

In an embodiment of the present disclosure~~invention~~, the composition comprises one or more preservatives. In a preferred embodiment, the one or more preservatives are selected from the group consisting of phenol, meta-cresol, and benzyl alcohol. Preferably, the preservative is meta-cresol. In an embodiment, the meta-cresol concentration is from about 2.5 mg/mL to about 3.8 mg/mL. Preferably, the meta-cresol concentration is about 3.15 mg/mL.

In an embodiment of the present disclosure~~invention~~, the composition comprises a tonicity agent. In an embodiment, the tonicity agent is selected from the group consisting of glycerol and sodium chloride. In a preferred embodiment, the tonicity agent is glycerol. In another preferred embodiment, the glycerol concentration is from about 5 to about 20 mg/mL. Preferably, the glycerol concentration is about 16 mg/mL.

In an embodiment, the composition may further comprise one or more stabilizing agents. In a preferred embodiment, the one or more stabilizing agents are selected from the group consisting of zinc, sodium chloride, calcium chloride, and arginine. Preferably, the stabilizing agent is zinc. Zinc oxide may be provided in the compositions in an amount sufficient to provide the desired number of zinc atoms. The compositions of certain embodiments of the present disclosure ~~invention~~ include zinc at a ratio of about 2-4 zinc atoms per six molecules of insulin. Certain preferred compositions will have about 2.5-3.5 atoms of zinc per six molecules of insulin. In certain embodiments, the concentration of zinc is from about 0.00525 mg/mL to about 0.131 mg/mL. Preferably, when the insulin concentration is about 100 IU/mL, the zinc concentration is about 0.0197 mg/mL.

In an embodiment, the composition may further comprise a buffering agent. In certain embodiments, the buffering agent is selected from the group consisting of

phosphate buffers, such as dibasic sodium phosphate, TRIS (2-amino-2-hydroxymethyl-propane-1,3-diol; Tromethamine) or sodium acetate. In certain preferred embodiments, the buffering agent is phosphate or TRIS.

In certain embodiments of the ~~invention~~present disclosure, the pH of the composition is about 7.0 to about 7.8. Preferably, the pH is about 7.4.

In an embodiment, the composition comprises insulin lispro in a concentration from about 40 to about 500 IU/mL, treprostinil in a concentration from about 0.01 to about 30 μM, meta-cresol in a concentration from about 2.5 to about 3.8 mg/mL, glycerol in a concentration from about 5 to about 20 mg/mL, zinc in a concentration from about 0.00525 to about 0.131 mg/mL, and the pH of the composition is from about 7.0 to about 7.8. In a preferred embodiment, the insulin concentration is about 100 IU/mL, treprostinil concentration is from about 0.05 to about 26 μM, zinc concentration is about 0.0197 mg/mL, meta-cresol concentration is about 3.15 mg/mL, glycerol concentration is about 16 mg/mL, and the pH of the composition is about 7.4.

The ~~According to a second aspect of the~~ present disclosure also ~~invention, there is~~ provide~~s~~d a method of treating diabetes comprising administering to a human in need thereof an effective dose of any of the aforementioned compositions.

The~~According to a third aspect of the~~ present disclosure further ~~invention, there is~~ provide~~s~~d any of the aforementioned compositions for use as a medicament. In an embodiment, the use of the composition is for use in the treatment of diabetes. The present disclosure ~~invention~~ also provides the use of any of the aforementioned compositions for the manufacture of a medicament for the treatment of diabetes.

The ~~According to a fourth aspect of the~~ present disclosure also ~~invention, there is~~ provide~~s~~d an article of manufacture comprising any of the aforementioned compositions. In an embodiment, the article of manufacture is a multi-use vial. In another embodiment, the article of manufacture is a re-usable pen injector. In another embodiment, the article of manufacture is a pump device for continuous subcutaneous insulin infusion therapy.

The ~~In another aspect, the~~ present disclosure further ~~invention~~ provides for insulin for use in combination with treprostinil. The ~~In another aspect, the~~ present disclosure also ~~invention~~ provides for treprostinil for use in combination with insulin.

The onset of action and/or absorption profile for an insulin product can be measured in a number of ways. For example, serum insulin concentrations can be

measured to calculate pharmacokinetic parameters relevant to time action, such as the time to maximum insulin concentration (Tmax), as well as the maximum insulin concentration (Cmax), and the time to reach the total insulin exposure, which is measured as the total area under the insulin concentration curve from 0 to infinity (AUCinf). Similarly, the glucose infusion rate (GIR) required to maintain normal serum glucose concentrations after insulin administration provides a direct measure of the pharmacodynamic action of the insulin. The present disclosure ~~invention~~ provides for a more rapid uptake of insulin into the blood and/or more rapid onset of action than existing commercial insulin products as reflected, for example, in shorter Tmax, shorter time to reach 50% of the Cmax (early 50% Tmax), shorter time for 50% of the Cmax to be cleared (late 50% Tmax), shorter time to reach 10%, 20% or 50% of the AUCinf, shorter time to maximal GIR or 50% maximal GIR, and/or other pharmacokinetic and/or pharmacodynamic parameters that would reflect more rapid absorption or action of the insulin.

When used herein, the term "composition" refers to a fixed dose combination of insulin and treprostinil, wherein the insulin, treprostinil, and any other ingredients or excipients, are combined in a single combined formulation.

When used herein, "insulin" means human insulin or a rapid-acting structural variant, mutant, or analog of human insulin that has the functional activity of but faster onset of action than human insulin. Particular rapid-acting analogs of human insulin are insulin lispro, insulin aspart, and insulin glulisine. Insulin for commercial products may be produced using recombinant DNA methods or by chemical synthesis. Recombinant methods are well-known and are strongly preferred. A molecule of human insulin (CAS No. 11061-68-0) consists of two amino acid chains, A and B, whose sequences are well-known.

The human insulin A-chain has the following sequence of amino acids:
Gly Ile Val Glu Gln Cys Cys Thr Ser Ile Cys Ser Leu Tyr Gln Leu Glu Asn Tyr Cys Asn (SEQ ID NO: 1).

The human insulin B-chain has the following sequence of amino acids:
Phe Val Asn Gln His Leu Cys Gly Ser His Leu Val Glu Ala Leu Tyr Leu Val Cys Gly Glu Arg Gly Phe Phe Tyr Thr Pro Lys Thr (SEQ ID NO: 2).