Artificial tears comprising sodium hyaluronate and carboxymethylcellulose

Figure 1

Embodiments described herein relate to formulations for and methods of use for eye drop formulations comprising carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.
By Inventors: Bereth J. Beard, Wendy Blanda, David Marsh, Peter Simmons, Joseph G. Vehige, Haixia Liu and Steven Matsumoto

RELATED PATENT DOCUMENTS
This application claims the benefit of United States Provisional Patent Application Serial No. 61/785,857, filed March 14, 2013, and United States Provisional Patent Application Serial No. 61/759,710, filed February 1, 2013, all of which are hereby incorporated by reference in their entirety.

FIELD OF THE INVENTION
Embodiments described herein relate to formulations for and methods of use for eye drop formulations comprising Carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

SUMMARY OF THE RELATED ART
Carboxymethyl cellulose (CMC) and hyaluronic acid (HA) are both compounds that may be used in commercial ophthalmic formulations.

United States Patent 5017229 "Water Insoluble Derivatives of Hyaluronic Acid" discusses a water insoluble biocompatible gel that includes the reaction product of hyaluronic acid, a polyanionic polysaccharide, and an activating agent. CMC is one of the polyanionic polysaccharides claimed. The product claimed is not an eye drop, but a film or a gel.

United States Patent Application 2010/0086512 "Mucomimetic compositions and uses thereof" discloses an eye drop comprising of a cationic antimicrobial agent with a magnesium calcium or magnesium/calcium complex of an anionic polymer such as HA and/or CMC. The patent application claims muco adhesion and maintained efficacy of the antimicrobial agent.
United States Patent 6472379, "Angiogenesis inhibition", discloses a formulation that inhibits angiogenesis and contains HA, CMC, and carbodiimide as a film or as a gel.

In the Asian Journal of Surgery, Volume 33, Issue 1, January 2010, pages 25-30, Yoo Seung Chung et al discuss "Anti-adhesive effect and safety of sodium hyaluronate and sodium carboxymethyl cellulose solution in thyroid surgery". Post-thyroidectomy adhesion was not decreased by using a HA-CMC combination solution.

In the Journal of Cornea and External Disease, Ji Hwan Lee et al discuss "Efficacy of sodium hyaluronate and carboxymethylcellulose in treating mild to moderate dry eye disease". A clinical study is discussed where patients are given either 0.1% sodium hyaluronate solution or 0.5% CMC solution. However, the study does not discuss the combination of HA and CMC together.

Genzyme, Inc., produces an anti-adhesion film called Seprafilm. The film is made of HA and CMC.

**BRIEF SUMMARY OF THE INVENTION**

The present invention relates to an eye drop formulation comprising carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

In another aspect, the present invention relates to a method comprising administering to a person suffering from Dry Eye Syndrome, an effective amount of a composition comprising carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

In another aspect, the present invention relates to a method comprising administering to a person suffering from Dry Eye Syndrome, an effective
amount of a composition comprising carboxymethyl cellulose (CMC) and hyaluronic acid (HA), glycerin, Purite®, boric acid, sodium borate decahydrate, potassium chloride, calcium chloride dehydrate, magnesium chloride hexahydrate, erythritol, sodium hydroxide, hydrochloric acid with an improved distribution on the cornea during blinking.

In another aspect, the present invention relates to artificial tears suitable for treating dry eye syndrome in a human or other mammal which comprises a mixture of carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

In one embodiment, a composition useful as an artificial tear comprises a mixture of carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking. In some embodiments, the composition may further comprise an alpha-hydroxyl acid (AHA). In some embodiments, the AHA may be lactic acid or lactate.

In some embodiments, the said mixture comprises from about 0.1 % to about 1.0% carboxymethyl cellulose (CMC). In some embodiments, the mixture comprises from about 0.05% to about 0.15% hyaluronic acid (HA). In some embodiments, the mixture comprises 0.5% carboxymethyl cellulose (CMC) and 0.1 % hyaluronic acid. The composition may further comprise glycerin. The composition may further comprise boric acid. The composition may further comprise sodium borate decahydrate. The composition may further comprise sodium citrate dihydrate. The composition may further comprise sodium lactate. The composition may further comprise potassium chloride. The composition may further comprise calcium chloride dehydrate. The composition magnesium chloride hexahydrate. The composition may further comprise erythritol. The composition may further comprise levocarnitine. The composition may further comprise sodium hydroxide. The composition may further comprise hydrochloric acid. Preferably, the composition may further comprise purified water.
In another embodiment, a method of improving the visual acuity of a person in need thereof comprises topically administering to said person, in an effective amount, an ophthalmic composition comprising a mixture of carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking. Preferably, said person suffers from Dry Eye Syndrome. In some embodiments, the composition may further comprise an alpha-hydroxyl acid (AHA). In some embodiments, the AHA may be lactic acid or lactate.

In yet another embodiment, a composition useful as an artificial tear comprises about from about 0.5 to about 1% w/v carboxymethyl cellulose sodium, about 0.1 to about 0.15% w/v sodium hyaluronate, at least one buffering agent, at least one excipient, at least one pH adjuster, and water to balance.

In one embodiment, the at least one buffering agent is selected from the group consisting of boric acid, sodium borate decahydrate, sodium citrate dehydrate, and sodium lactate. In one embodiment, the at least one excipient is selected from the group consisting of potassium chloride, calcium chloride dihydrate, magnesium chloride hexahydrate, and erythritol. In one embodiment, the at least one pH adjuster is selected from the group consisting of sodium hydroxide and hydrochloric acid. The composition may further comprise about 0.5-1.0% w/v of glycerin. The composition may further comprise levocarnitine.

In some embodiments, the composition may comprise about 0.5% w/v carboxymethyl cellulose sodium, about 0.9% w/v glycerin, about 0.1% w/v sodium hyaluronate, about 0.01% w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1% w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, sufficient sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance. The composition may further
comprise about 0.5% w/v carboxymethyl cellulose sodium, about 0.9% w/v glycerin, about 0.15% w/v sodium hyaluronate, about 0.01% w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1% w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.006% w/v sodium lactate, about 0.1% w/v sodium hyaluronate, about 0.01% w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1% w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, about 0.25% levocarnitine, sufficient sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance. In yet another embodiment, the composition may comprise about 0.5% w/v carboxymethyl cellulose sodium, about 0.9% w/v glycerin, about 0.10% w/v sodium hyaluronate, about 0.01% w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1% w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, about 0.25% levocarnitine, sufficient sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance.

It will of course be understood that the ranges described above and throughout, are also intended to encompass single values contained within these ranges. For example, for a formulation comprising a particular ingredient in a range between 1-50%, a percentage of, for example, 5% or 49% is also intended to be disclosed.

One advantage of CMC is that ocular to systemic ratios are improved with viscous CMC formulations. One advantage of HA is that the shear dependent viscosity of the HA gives an improved distribution on the cornea during blinking.
We claim an unexpected result with the combination of CMC with HA in an eye drop.

There is an increase in viscosity which is observed when 0.5% carboxymethyl cellulose (CMC) is mixed with 0.1% hyaluronic Acid (HA). The increase in viscosity is more than additive which is unexpected. Therefore, we claim the discovery of a new eye drop formulation which has enhanced properties in comparison to formulations with single components. These enhanced properties are greater than would be theoretically expected.

**BRIEF DESCRIPTION OF THE DRAWINGS**

**Figure 1** describes the viscosity curves determined for CMC in PBS, HA in PBS, CMC with HA measured and CMC with HA expected.

**Figure 2** describes the Brookfield viscosity at 60 RPM determined for CMC in PBS, HA in PBS, CMC with HA measured and CMC with HA expected.

**Figures 3A-l** describe clinical study results and scoring for tested formulations in various categories pursuant to a study described in greater detail below during the length of the study.

**Figure 4** describes the change from baseline in tear breakup time for tested formulations pursuant to a study described in greater detail below during the length of the study.

**Figure 5** illustrates the change in baseline pursuant to a study described in greater detail below for combined corneal and conjunctival staining in tested formulations during the length of the study.

**Figure 6** illustrates the change in baseline pursuant to a study described in greater detail below for combined corneal and conjunctival staining in tested formulations during the length of the study for a subgroup showing clinically-important staining.

**Figure 7** illustrates the change in baseline pursuant to a study described in greater detail below for corneal staining in tested formulations during the length of the study for a subgroup showing clinically-important staining.
Figure 8 illustrates the change in baseline pursuant to a study described in greater detail below for corneal staining in tested formulations during the length of the study.

Figure 9 illustrates the change in ocular burning and stinging during the length of a study described in greater detail below.

Figure 10 illustrates the change in ocular dryness during the length of a study described in greater detail below.

Figure 11 illustrates the change in lid wiper epitheliopathy during the length of a study described in greater detail below.

DETAILED DESCRIPTION OF THE INVENTION

In one aspect of the invention, there is provided an eye drop formulation comprising Carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

One advantage of CMC is that ocular to systemic ratios are improved with viscous CMC formulations. One advantage of HA is that the shear dependent viscosity of the HA gives an improved distribution on the cornea during blinking. We claim an unexpected result with the combination of CMC with HA in an eye drop.

There is an increase in viscosity which is observed when about 0.1 % to about 1.0% Carboxymethyl cellulose (CMC) is mixed with about 0.05% to about 0.15% Hyaluronic Acid (HA). There is an increase in viscosity which is observed when about 0.5% Carboxymethyl cellulose (CMC) is mixed with about 0.1 % Hyaluronic Acid (HA). There is an increase in viscosity which is observed when 0.5% Carboxymethyl cellulose (CMC) is mixed with a 0.1 % Hyaluronic Acid (HA). The increase in viscosity is more than additive which is unexpected. Therefore, we claim the discovery of a new eye drop formulation which has enhanced properties in comparison to formulations with single components. These enhanced properties are greater than would be theoretically expected.
The following Table lists some examples of preferred formulations.

Table 1: Formulations

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Formulation 1: Concentration, % (w/v)</th>
<th>Formulation 2: Concentration, % (w/v)</th>
<th>Formulation 3: Concentration, % (w/v)</th>
<th>Role</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carboxymethyl cellulose sodium</td>
<td>0.5 % w/v</td>
<td>0.5 % w/v</td>
<td>0.5 % w/v</td>
<td>Active</td>
</tr>
<tr>
<td>Glycerin</td>
<td>0.9 % w/v</td>
<td>0.9 % w/v</td>
<td>0.9 % w/v</td>
<td>Active</td>
</tr>
<tr>
<td>Sodium hyaluronate</td>
<td>0.1 % w/v</td>
<td>0.15 % w/v</td>
<td>0.10 % w/v</td>
<td>Viscosity adj.</td>
</tr>
<tr>
<td>Purite®</td>
<td>0.01 % w/v</td>
<td>0.01 % w/v</td>
<td>0.01 % w/v</td>
<td>Preservative</td>
</tr>
<tr>
<td>Boric acid</td>
<td>0.7 % w/v</td>
<td>0.7 % w/v</td>
<td>0.7 % w/v</td>
<td>Buffering agent</td>
</tr>
<tr>
<td>Sodium borate decahydrate</td>
<td>0.2 % w/v</td>
<td>0.2 % w/v</td>
<td>0.2 % w/v</td>
<td>Buffering agent</td>
</tr>
<tr>
<td>Sodium citrate dihydrate</td>
<td>0.1 % w/v</td>
<td>0.1 % w/v</td>
<td>0.1 % w/v</td>
<td>Buffering agent</td>
</tr>
<tr>
<td>Potassium chloride</td>
<td>0.14 % w/v</td>
<td>0.14 % w/v</td>
<td>0.14 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Calcium chloride dihydrate</td>
<td>0.006 % w/v</td>
<td>0.006 % w/v</td>
<td>0.006 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Magnesium chloride hexahydrate</td>
<td>0.006 % w/v</td>
<td>0.006 % w/v</td>
<td>0.006 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Erythritol</td>
<td>0.5 % w/v</td>
<td>0.5 % w/v</td>
<td>0.25 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Levocarnitine</td>
<td>--</td>
<td>--</td>
<td>0.25 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Sodium hydroxide</td>
<td>7.2 pH</td>
<td>7.2 pH</td>
<td>7.2 pH</td>
<td>pH Adjust</td>
</tr>
<tr>
<td>Hydrochloric acid</td>
<td>7.2 pH</td>
<td>7.2 pH</td>
<td>7.2 pH</td>
<td>pH Adjust</td>
</tr>
<tr>
<td>Water for injection/purified water</td>
<td>100 % w/v</td>
<td>100 % w/v</td>
<td>100 % w/v</td>
<td>QS Adjust</td>
</tr>
</tbody>
</table>

With this in mind, embodiments described herein are not restricted to the percentages listed above. The amount of CMC may range between 0.01 % w/v to 10% w/v, preferably 0.3% w/v to 5% w/v, more preferably 0.5% w/v to 1%w/v, and most preferably 0.5%w/v. Other preferred percentages include 0.1 % w/v, 0.15%w/v, 0.2% w/v, 0.3% w/v, 0.4%w/v, 0.5% w/v, 0.6% w/v, 0.7% w/v, 0.8% w/v, 0.9%w/v, 1.0 %w/v, 1.5%w/v, 2%w/v, 3%w/v, 3.5%w/v, 4%w/v, and 5%w/v. The amount of HA may range between 0.01 % w/v to 10% w/v, preferably 0.05%w/v to 4%w/v, more preferably 0.1 %w/v to 1%w/v, and most preferably 0.1 %w/v to 0.15%w/v. Other preferred percentages include
0.01%w/v, 0.02%w/v, 0.03%w/v, 0.04%w/v, 0.05%w/v, 0.06%w/v, 0.07%w/v, 0.08%w/v, 0.09%w/v, 0.1%w/v, 0.125%w/v, 0.15%w/v, 0.2%w/v, 0.3%w/v, 0.4%w/v, 0.5%w/v, 0.6%w/v, 0.7%w/v, 0.8%w/v, 0.9%w/v, 1.0%w/v, 1.5%w/v, 2%w/v, 3%w/v, 3.5%w/v, 4%w/v, and 5%w/v.

The carboxymethyl cellulose used in embodiments described herein is preferably of any grade that can be formulated for topical use. Preferably, the carboxymethyl cellulose is sodium carboxymethyl cellulose. More preferably, the carboxymethyl cellulose is sodium carboxymethyl cellulose with a low viscosity.

The HA used in embodiments described herein is preferably of any grade that can be formulated for topical use. Preferably, the hyaluronic acid is in the form of sodium hyaluronate. In some embodiments, the hyaluronic acid has an intrinsic viscosity of about 0.5 m³/kg to about 4.0 m³/kg, more preferably about 1.1 m³/kg to about 2.0 m³/kg, or also more preferably about 2.5 m³/kg to about 4.0 m³/kg. Some embodiments may have an intrinsic viscosity of about 2.2 m³/kg to about 2.6 m³/kg, and some embodiments may have an intrinsic viscosity of about 1.1 m³/kg to about 3.0 m³/kg.

The above formulations are intended to be examples of possible formulations, and different formulations, especially in terms of the excipients and other ingredients, are of course possible. For example, it is within the scope of the embodiments disclosed herein that some formulations may omit Purite. In some embodiments, Purite may be substituted for or combined with another preservative. In some embodiments, the formulations may be unpreserved. As shown in Formulation 3, it is possible for additional excipients to be added to certain embodiments, and formulations may contain carnitine and its isomers (for example, L-carnitine or levocarnitine). In some embodiments, carnitine may be present from about 0.01%w/v to about 1%w/v, preferably from about 0.1%w/v to about 0.5%w/v, and most preferably at 0.25%w/v.
This formulation demonstrates a marked drop in viscosity upon application of a shear force; the use of a wetting drop with a high shear drop in viscosity suggests that, when the formulation is dropped in the eye, the eyelid will produce sufficient shear-force to produce a low-viscosity, comfortable, eye drop.

But, once in the cul-de-sac (i.e., an eye corner) where there is no shear, the formulation should rapidly become more viscous. Having this "reservoir" of viscous polymers will maintain a sustained level of polymers in the tear fluid. The polymers in the tear fluid will limit the evaporation of tears and, in this way, counter dry eye. Concomitantly, the sheer force of the eyelid should prevent the polymers from becoming too viscous and blurring vision.

In addition, the viscous material in the cul-de-sac (e.g., in the puncta and in the canaliculus) slows the drain of tears from the eye and therefore, has a second action to treat dry eye by building tear fluid up in front of the eye.

In some embodiments, compositions and methods directed to eye drop compositions may include an alpha-hydroxyl acid ("AHA"). Preferably, the AHA is lactic acid, or lactate. Other AHAs include glycolic acid.

It has been found that AHA-containing compositions and formulations may benefit from the inclusion of an AHA, as cells—such as cells already damaged by dry eye and other ocular conditions—can be gently and selectively exfoliated by an AHA. Accordingly, some embodiments may comprise the administration of an AHA-containing formulation that also comprises compatible solutes. Without wishing to be bound by theory, it is believed that such formulations may protect cells (e.g., ocular and conjunctival cells) from stress and facilitate cell renewal. This may increase the overall ocular health of the eye to which the formulation is applied.
A normal ocular surface is covered with non-keratinized corneal and conjunctival epithelial cells. The non-keratinized superficial epithelial cells produce bound mucins (glycocalyx) that coat the ocular surface to create a hydrophilic surface that stabilizes the tear film. Goblet cells are interspersed between conjunctival epithelial cells, and secret soluble unbound mucins that stabilize the tear film by reducing surface tension, as well as lubricate and protect the ocular surface. For the special structure and function of ocular surface, it is most preferable to keep the epithelial cells non-keratinized to maintain its health and integrity.

However, in an adverse condition like dry eye, the ocular surface integrity may be disrupted due to the stress of hyperosmolarity and/or desiccation. Again, without wishing to be bound by theory, some studies have shown that hyperosmotic stress can activate the mitogen-activated protein kinase signaling pathway, which further leads to squamous metaplasia, or "cornification". Cornification disrupts the corneal epithelial barrier function. The cornified epithelial cells may then lose their glycocalyx coverage and become poorly hydrated, thereby destabilizing the tear film and exacerbating ocular desiccation. Some research has demonstrated that the cornified conjunctival epithelial cells can entrap the conjunctival goblet cells, blocking their mucus secretion, and further degrading the tear quality and stability.

Accordingly, it would be advantageous to provide an AHA-containing formulation that protects ocular epithelial cells from stress and/or cornification. Also, it would be advantageous to provide an AHA-containing formulation that facilitates the shedding of cornified cells, thereby improving the renewal of the ocular surface.

Preferably, the AHA-containing formulation comprises one or more solute components, for example, one or more solute components selected from the group consisting of carnitine (including levocarnitine), betaine, glycerin, and erythritol. Some studies have shown that these small molecules may protect
cells from hyperosmotic stress. Again, without wishing to be bound by theory, it is believed that these solute components may enter and accumulate in cells, thereby balancing the osmotic difference with the extracellular fluid. Some studies on the cultured corneal epithelial cells have found that these and similar solutes significantly lowered the levels of MAPK activation in response to hyperosmotic stress. As it is believed that cornification may be mediated by MAPK pathway, compatible solutes may prevent cell cornification, as well as inflammation and other damage.

AHA, including lactic acid and glycolic acid, has been widely used in skin care as a gentle exfoliator at low concentration. AHA is believed to selectively affect epithelial keratinization, thereby diminishing the cellular cohesion between corneocytes at the lowest levels of the stratum corneum. This action promotes exfoliation of the outermost layer of the skin. Especially when used in high concentration and low pH, AHA can produce a rapid loss of skin cells, as seen in a "chemical peel." However, the exfoliating effect of AHA is not solely attributable to its low pH, as many AHA containing skin care products have a pH of near neutral with a gentle but efficient exfoliating effect.

Accordingly, in one embodiment, a preferred composition comprises a lactate buffer. Preferably, this lactate buffer composition is maintained a neutral or near-neutral pH. This composition may be beneficial in promoting the shedding of cornified epithelial cells so as to maintain ocular surface integrity. The removal of the cornified cells may also eliminate the formation of dry spots on the ocular surface and enhance tear film distribution over the ocular surface. In addition, removal of cornified epithelia may open up the entrapped goblet cells and help the recovery of goblet cell function to allow better delivery of mucins to the ocular surface. The combination of the two effects may further be beneficial in stabilizing the tear film and in protecting the ocular surface. The lactate buffer may also be more biocompatible with other commonly-used buffers, as lactate acid is a by-product of glucose metabolism that naturally exists in the tears. Studies have shown that lactate may participate in wound
healing, stimulating collagen and hyaluronan synthesis. In addition, as a small solute, lactate may also serve as an intra-cellular osmolyte that protects the ocular surface from hyperosmotic stress by a mechanism similar to or synergistic with the osmoprotectant compatible solutes. The potential osmoprotection of lactate may help further reduce cornification.

Compositions containing AHA, preferably lactic acid or lactate, may be formulated in accordance with the other embodiments described herein. For example, a preferred formulation may be adapted from the example formulations described in Table 1. Further, a preferred formulation may be adapted from the Table below.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Formulation 4: Concentration, % (w/v)</th>
<th>Role</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carboxymethylcellulose sodium</td>
<td>0.5 % w/v</td>
<td>Active</td>
</tr>
<tr>
<td>Glycerin</td>
<td>1.0 % w/v</td>
<td>Active</td>
</tr>
<tr>
<td>Sodium hyaluronate</td>
<td>0.1 % w/v</td>
<td>Viscosity adj.</td>
</tr>
<tr>
<td>Sodium lactate</td>
<td>0.3% w/v</td>
<td>Buffering agent</td>
</tr>
<tr>
<td>Potassium chloride</td>
<td>0.14% w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Calcium chloride dihydrate</td>
<td>0.006 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Magnesium chloride hexahydrate</td>
<td>0.006 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Erythritol</td>
<td>0.5 % w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Levocarnitine</td>
<td>0.25% w/v</td>
<td>Excipient</td>
</tr>
<tr>
<td>Sodium hydroxide</td>
<td>6.2-7.0 pH</td>
<td>pH Adjust</td>
</tr>
<tr>
<td>Hydrochloric acid</td>
<td>6.2-7.0 pH</td>
<td>pH Adjust</td>
</tr>
<tr>
<td>Water for injection/purified water</td>
<td>100 % w/v</td>
<td>QS Adjust</td>
</tr>
</tbody>
</table>

With this in mind, embodiments described herein are not restricted to the percentages listed above. The amount of CMC may range between 0.01 % w/v to 10% w/v, preferably 0.3% w/v to 5% w/v, more preferably 0.5% w/v to 1%w/v, and most preferably 0.5%w/v. Other preferred percentages include
0.1% w/v, 0.15% w/v, 0.2% w/v, 0.3% w/v, 0.4% w/v, 0.5% w/v, 0.6% w/v, 0.7% w/v, 0.8% w/v, 0.9% w/v, 1.0% w/v, 1.5% w/v, 2% w/v, 3% w/v, 3.5% w/v, 4% w/v, and 5% w/v. The amount of HA may range between 0.01% w/v to 10% w/v, preferably 0.05% w/v to 4% w/v, more preferably 0.1% w/v to 1% w/v, and most preferably 0.1% w/v to 0.15% w/v. Other preferred percentages include 0.01% w/v, 0.02% w/v, 0.03% w/v, 0.04% w/v, 0.05% w/v, 0.06% w/v, 0.07% w/v, 0.08% w/v, 0.09% w/v, 0.1% w/v, 0.125% w/v, 0.15% w/v, 0.2% w/v, 0.3% w/v, 0.4% w/v, 0.5% w/v, 0.6% w/v, 0.7% w/v, 0.8% w/v, 0.9% w/v, 1.0% w/v, 1.5% w/v, 2% w/v, 3% w/v, 3.5% w/v, 4% w/v, and 5% w/v.

The carboxymethyl cellulose used in embodiments described herein is preferably of any grade that can be formulated for topical use. Preferably, the carboxymethyl cellulose is sodium carboxymethyl cellulose. More preferably, the carboxymethyl cellulose is sodium carboxymethyl cellulose with a low viscosity.

The HA used in embodiments described herein is preferably of any grade that can be formulated for topical use. Preferably, the hyaluronic acid is in the form of sodium hyaluronate. In some embodiments, the hyaluronic acid has an intrinsic viscosity of about 0.5 m³/kg to about 4.0 m³/kg, more preferably about 0.9 m³/kg to about 3.0 m³/kg. In some preferred embodiments, the hyaluronic acid has an average molecular weight from about 2.0 to about 2.6 million Daltons. In some other preferred embodiments, the hyaluronic acid has an intrinsic viscosity from about 1.1 m³/kg to about 2.0 nV/kg. In some other preferred embodiments, the hyaluronic acid has an average molecular weight from about 0.5 to about 1.2 million Daltons.

The amounts of glycerin used in the embodiments described herein may range from about 0.5% w/v to about 2% w/v, and is preferably from 0.9% w/v to about 1.5% w/v, and most preferably 0.9% w/v or 1.0% w/v. The lactate/lactic acid buffering agent may use any suitable lactate salt, most preferably sodium lactate. It may be used at a concentration between 0.1% w/v to about 1.0% w/v.
w/v, most preferably 0.3% w/v. In some embodiments, the lactate buffer may be combined with other buffering agents. Some embodiments may further comprise one or more preservatives, such as benzalkonium chloride or Purite®.

The formulations described in Tables 1 and 2 herein are not intended to be limiting, and combinations and modifications to these formulations may be made. For example, some embodiments may combine one or more buffering agents, such as boric acid and sodium lactate.

**Example 1**

In one test, shown below, 0.5% CMC and 0.1 % HA solutions were tested for viscosity in comparison to a solution that contained both 0.5% CMC and 0.1 % HA (Formulation 1 in the table above). At several different frequencies tested, the viscosity of the combination formulation was greater than the predicted value (calculated by addition of the values for the individual formulations). This shows that the desired viscosity can be obtained by using less CMC and/or HA than one would predict based, if a combination is utilized instead of a single polymer.

As stated above, the shear thinning characteristic is a desirable characteristic of HA formulations. However, HA is quite expensive in comparison to CMC. In the example where 0.5% CMC and 0.1 % HA solutions were tested for viscosity in comparison to a solution that contained both 0.5% CMC and 0.1 % HA, shear thinning can be quantified by taking the ratio of the viscosity, in centipoise (cps), at 1/s to the viscosity at 10/s. These results are summarized in Table 3 below, as well as Figure 1. Figure 2 illustrates the viscosity in cps at 60rpm.
Although there is more CMC than HA in the formulation, the shear thinning ratio of the combination is high - close to that of the HA alone. This shows that a CMC formulation can be made to shear thin with the addition of HA.

At this time, it is not known if the 0.5% CMC to 0.1 % HA is the optimal ratio of these polymers. An even greater gain in viscosity and enhanced shear thinning might be realized by a slightly different combination of CMC and HA.

Without wishing to be bound by theory, it is possible that the unexpected increase in viscosity demonstrates that the polymers have an unexpected positive interaction. This interaction is unlikely to be charge-charge interaction since both sodium hyaluronate and sodium carboxymethyl cellulose are slightly negatively charged when in solution. The interaction is likely chain entanglement, which can have a positive clinical benefit. It is theorized, clinically, that CMC may have more binding force to corneal cells while HA is a better lubricant but has less ocular surface retention. In combination, the CMC assists in HA retention, via entanglement, to the cell membrane.

<table>
<thead>
<tr>
<th>Table 3: Shear thinning</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td>Formulation</td>
</tr>
<tr>
<td>0.5% CMC in PBS</td>
</tr>
<tr>
<td>0.1 % HA in PBS</td>
</tr>
<tr>
<td>Predicted</td>
</tr>
<tr>
<td>0.5% CMC + 0.1% HA</td>
</tr>
<tr>
<td>Actual</td>
</tr>
<tr>
<td>0.5% CMC + 0.1% HA</td>
</tr>
</tbody>
</table>
The HA-CMC-cornea attraction gives a better, more durable lubricant system. In addition, since both polymers have been shown to contribute to cell migration in vitro, there could be a combined enhanced benefit in vivo.

**Example 2**

Testing was performed on human volunteers, using two CMC-HA formulations. The first formulation used a 0.5% CMC/0.1% HA formulation ("EDNP-1") as set forth above as Formulation 1 in Table 1. The second formulation used a 0.5% CMC/0.15% HA formulation ("EDNP-2") as set forth above as Formulation 2 in Table 1. The two Formulations were compared to an existing product known to be effective in treating dry eye—Refresh® Tears with 0.5% CMC, salts, and Purite® ("Refresh"). It is important to note that the Refresh product used in testing does not, strictly speaking, represent a control with little therapeutic activity (e.g., pure saline). Rather, the Refresh product represents an established, successful product that is known to be efficacious in treating dry eye.

Approximately 100 individuals were in each treatment group, with dosing done as needed, and at least twice daily, for 90 days. The primary endpoint was a change in the Ocular Surface Disease Index (OSDI) at day 90 of the testing. The OSDI is a 12-question standardized test that surveys patients on the existence and severity of various dry-eye related attributes, including light sensitivity, perceived eye grittiness, vision blurring, and eye comfort at various times and when performing various activities.

The study population consisted of current eye drop users with mild to severe symptoms and signs of dry eye, so as to obtain a generally heterogeneous population that would provide a good indication of the efficacy of the two tested Formulations. The population therefore presented with an OSDI between 18 and 65, a tear breakup time (TBUT) of less than 10 seconds, and presenting some corneal and/or conjunctival surface staining. Patients showing especially severe dry eye were excluded.
Figures 3A-F depict the average scoring in specific OSDI sub-categories for the tested formulations. In Figure 3A, OSDI ocular symptoms for EDNP-1 appeared to be improved over Formulations 2 and the Refresh product. This is evidenced by the lower average OSDI score (or improvement from baseline) at the end of the 90-day period. Figure 3B, showing the median OSDI ocular symptom score, also shows that EDNP-1 has a greater median improvement compared to the other Formulations.

Figure 3C shows that the average OSDI visual function score for EDNP-1 also demonstrates the greatest level of improvement compared to EDNP-2 and Refresh by the end of the 90-day period. In Figure 3D, the median OSDI visual function score shows that EDNP-1 is slightly better than Refresh.

Figure 3E illustrates that the average OSDI score for environmental triggers also shows the most improvement for EDNP-1 compared to EDNP-2 and Refresh by the end of the 90-day period. The median score illustrated in Figure 3F shows that EDNP-1 and -2 are both superior to Refresh.

Figure 3G looks specifically to the average dryness symptom score for perceived ocular dryness during the length of the study. Here, while all compositions registered some improvement over the baseline, EDNP-1 showed the greatest improvement over the study duration.

Figure 3H illustrates the results of administering EDNP-1 as compared to Refresh in a study subgroup suffering from severe dry eye, as evidenced by a combined corneal and conjunctival staining score greater than or equal to 15 (the relevance of staining to the severity of dry eye is discussed in greater detail below). Here, EDNP-1 performed better than Refresh at all followup visits from day 7 to day 90, as shown by the reduction in the OSDI score from baseline.
Figure 3 illustrates the change in staining score (again, discussed in greater detail below) for a study subgroup suffering from severe dry eye, based on a VAS eye dryness symptom scale greater than or equal to 66. The VAS eye dryness symptom scale rates the overall severity for ocular dryness in a subject over the course of a week, where a score of zero indicates no ocular dryness, to a maximum possible score of 100, indicating maximal dryness. The VAS eye dryness scale measures the single symptom of ocular dryness, in contrast with the OSDI score being a comprehensive assessment of dry eye symptoms including ocular discomfort, environmentally-triggered, and vision-related symptoms. Here, the EDNP-1 formulation performed better than Refresh at reducing corneal and conjunctival staining for the duration of the study.

Figure 4 is a graph showing the average change in tear break up time, in seconds, from the beginning of the study through 90 days. Here, EDNP-2 showed a greater average change in tear break up time, compared to EDNP-1 and Refresh.

Figure 5 shows the average change during the study from baseline of combined corneal and conjunctival staining in the entire treatment group. Generally, corneal and conjunctival staining is determined by administering a dye to the eye which is capable of staining the exposed ocular surfaces, such as fluorescein, rose bengal, and lissamine green. Staining is believed to indicate areas of the eye which have been damaged as a result of dry eye, and it has been postulated that the dyes stain ocular epithelial surfaces that are lacking a protective mucin protein coat or that otherwise present exposed, unprotected epithelial cell surfaces. Ocular staining can be scored based on the location and amount of staining on different sections and regions of the eye. For example, the cornea may be subdivided into several sections, and the amount of staining quantified and assigned a score. Similar scoring may be applied to the conjunctiva. Examples of such methodologies may be found, for example, in Foulks, "Challenges and Pitfalls in Clinical Trials of Treatments for Dry Eye," *Ocular Surface*, V. 1, N. 1, pp. 20-30 (2003); and Baudouin, et al,

In the present study, a modified NEI Grid was used with a total of 11 zones distributed throughout the eye. Scoring was assigned using a similar rationale to the Baudouin study cited above, where different ocular zones were scored in relation to the extent of staining, which therefore indicated the severity of the dry eye. Here, EDNP-1 and -2 were better than Refresh in reducing ocular staining, and in particular by the end of the study.

Turning now to Figure 6, average corneal and conjunctival staining scores were tracked during the length of the study for the clinically-important staining subgroup, and compared to the staining score at the beginning of the study. A clinically important staining score was set to be greater than or equal to 15, and the clinically-important staining subgroup was defined as such. This score was used as an indication of patients suffering from more severe dry eye, and the scoring indicates that at least a third of the 11 ocular zones would have a staining score greater than 1. Here, both EDNP-1 and -2 were significantly better than Refresh at reducing corneal and conjunctival staining in a study population with severe dry eye, as shown by the improvement (reduction) in staining scores.

Focusing on the extent of the corneal staining in the clinically important subgroup, Figure 7 shows the average improvement from baseline in this group during the length of the study. Specifically, by the endpoint of the study, EDNP-1 was found to be far superior to the other two formulations in reducing the extent of corneal staining, and as such dry eye, in the group demonstrating clinically-important ocular staining. EDNP-2 was also marginally better than Refresh.
Figure 8 shows a graph of the average corneal staining score change in all subjects, tracked during the length of the entire study. Here, both EDNP-1 and -2 were better than Refresh formulation, with EDNP-1 showing the greatest reduction in corneal staining from baseline as compared to the other tested substances. As such, the tested formulations provide a therapeutic benefit to the majority of study participants and not only the group showing clinically-important ocular staining.

**Example 3**

Another clinical test was performed on human volunteers. Here, 365 contact lens users were randomized in a 2:1 treatment allocation with stratification by 6 types of hydrogel, silicon hydrogel, and rigid gas-permeable contact lenses. They were instructed to instill 1 to 2 drops of the studied eye drops in each eye a minimum of 4 times per day for 90 days. One of these uses may be to prepare the contact lens for wear by placing 1 to 2 drops on the inner surface of the contact lens prior to insertion. In this study, Formulation 1 was compared to the Refresh Tears® ("Refresh") eye drops previously described.

The contact lens users were randomized into the study at fifteen different sites in the United States, with 350 subjects (95.9%) completing the study. There were no significant differences between treatment groups in regard to age, sex, or race, or in regard to the number of treatment-related discontinuations.

As shown in Figure 9, ocular symptoms of burning and stinging throughout the day were assessed on a VAS burning/stinging scale over a total period of 90 days. A statistically-significant improvement from baseline in users of Formulation 1 in the reduction of symptoms of burning and stinging was shown at 7, 30, and 90 days. Also, by the end of the 90 day period, there was a statistically significant difference and improvement in the reduction of burning and stinging symptoms by Formulation 1 as compared to Refresh.
With reference now to Figure 10, the treatment of the ocular symptom of dryness after use of Formulation 1 was compared to Refresh. Here, Formulation 1 showed a significant improvement from baseline dryness at 7, 30, 60, and 90 days from administration. Further, Formulation 1 showed a statistically significant improvement in the reduction of ocular dryness as compared to Refresh by at least days 60 and 90. This shows that Formulation 1 provides superior long-term reduction in ocular dryness.

Figure 11 compares the progression of lid wiper epitheliopathy (LWE) between Formulation 1 and Refresh. LWE is a disorder of the marginal conjunctiva of the upper eyelid with dry eye symptoms. LWE may be related to mechanical forces during blinking resulting in inflammation of the ocular surface. LWE was assessed in the study with lissamine green with Korb's Grading (Korb et al, 2010). The upper eyelid was everted with great care to avoid contact with the region of the lid wiper. White light of low-to-moderate intensity with diffuser is used for LWE observation, and LWE was graded from 0 (< 2 mm) to 3 (> 10 mm) based on the horizontal length involved and from 0 (< 25%) to 3 (> 75%) based on the average sagittal height involved. The individual grades for these 2 findings were averaged for a final lissamine green staining grade for the lid wiper of each eye. Here, Formulation 1 showed improvement in reducing LWE from baseline, and was significantly better than Refresh at the end of the 90 day period.

Accordingly, this study demonstrates that Formulation 1 is superior to Refresh at ameliorating and treating dry eye symptoms.

The present invention is not to be limited in scope by the exemplified embodiments, which are only intended as illustrations of specific aspects of the invention. Various modifications of the invention, in addition to those disclosed herein, will be apparent to those skilled in the art by a careful reading of the specification, including the claims, as originally filed. It is intended that all such modifications will fall within the scope of the appended claims.
What is claimed is:

1. A composition useful as an artificial tear, which comprises a mixture of Carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

2. The composition of claim 1 wherein said mixture comprises from about 0.1% to about 1.0% Carboxymethyl cellulose (CMC).

3. The composition of any of claims 1 to 2, wherein said mixture comprises from about 0.05% to about 0.15% hyaluronic acid (HA).

4. The composition of any preceding claim, wherein said mixture comprises 0.5% Carboxymethyl cellulose (CMC) and 0.1% Hyaluronic Acid.

5. The composition of any preceding claim, further comprising Glycerin.

6. The composition of any preceding claim, further comprising Boric acid.

7. The composition of any preceding claim, further comprising Sodium borate decahydrate.

8. The composition of any preceding claim, further comprising Sodium citrate dihydrate.

9. The composition of any preceding claim, further comprising sodium lactate.

10. The composition of any preceding claim, further comprising Potassium chloride.

11. The composition of any preceding claim, further comprising Calcium chloride hydrate.

12. The composition of any preceding claim, further comprising Magnesium chloride hexahydrate.

13. The composition of any preceding claim, further comprising Erythritol.

14. The composition of any preceding claim, further comprising levocarnitine.

15. The composition of any preceding claim, further comprising Sodium hydroxide.
16. The composition of any preceding claim, further comprising Hydrochloric acid.

17. The composition of any preceding claim, further comprising purified water.

18. The method of improving the visual acuity of a person in need thereof which comprises topically administering to said person, in an effective amount, an ophthalmic composition comprising a mixture of Carboxymethyl cellulose (CMC) and hyaluronic acid (HA) with an improved distribution on the cornea during blinking.

19. The method of claim 18 wherein said person suffers from Dry Eye Syndrome.

20. A composition useful as an artificial tear, the composition comprising from about 0.5% w/v to about 1% w/v carboxymethylcellulose sodium, about 0.1 to about 0.15% w/v sodium hyaluronate, at least one buffering agent, at least one excipient, at least one PH adjuster, and water to balance.

21. The composition of claim 20, wherein the at least one buffering agent is selected from the group consisting of boric acid, sodium borate decahydrate, sodium citrate dehydrate, and sodium lactate.

22. The composition of any of claims 20 to 21, wherein the at least one excipient is selected from the group consisting of potassium chloride, calcium chloride dihydrate, magnesium chloride hexahydrate, and erythritol.

23. The composition of any of claims 20-22, wherein the at least one pH adjuster is selected from the group consisting of sodium hydroxide and hydrochloric acid.

24. The composition of any of claims 20-23, wherein the composition further comprises about 0.5 to about 1.0% w/v of glycerin.

25. The composition of any of claims 20-24, wherein the composition further comprises levocarnitine.

26. The composition of any of claims 20-24, wherein the composition comprises about 0.5% w/v carboxymethylcellulose sodium, about 0.9%
glycerin, about 0.1 % w/v sodium hyaluronate, about 0.01 % w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1 % w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, sufficient sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance.

27. The composition of any of claims 20-24, wherein the composition comprises about 0.5% w/v carboxymethylcellulose sodium, about 0.9% w/v glycerin, about 0.15% w/v sodium hyaluronate, about 0.01 % w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1 % w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, sufficient sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance.

28. The composition of any of claims 20-25, wherein the composition comprises about 0.5% w/v carboxymethylcellulose sodium, about 0.9% w/v glycerin, about 0.10% w/v sodium hyaluronate, about 0.01 % w/v Purite®, about 0.7% w/v boric acid, about 0.2% w/v sodium borate decahydrate, about 0.1 % w/v sodium citrate dihydrate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, about 0.25% levocarnitine, sufficient sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance.

29. The composition of any of claims 20-25, wherein the composition comprises about 0.5% w/v carboxymethylcellulose sodium, about 1.0% w/v glycerin, about 0.10% w/v sodium hyaluronate, about 0.3% w/v sodium lactate, about 0.14% w/v potassium chloride, about 0.006% w/v calcium chloride dihydrate, about 0.006% w/v magnesium chloride hexahydrate, about 0.5% erythritol, about 0.25% levocarnitine, sufficient
sodium hydroxide and hydrochloric acid to adjust the composition pH to 7.2, and water to balance.
Figure 3F

Figure 3E
Symptom Scales

Dryness

Figure 3G
Change from Baseline in Tear Break-up Time (seconds) by Treatment
Change from Baseline in Combined Corneal and Conjunctival Staining by Treatment

Figure 5
Change from Baseline in Combined Corneal and Conjunctival Staining – Clinically Important Subgroup

Figure 6
Change from Baseline in Corneal Staining

Figure 8
Figure 9

Daytime Ocular Symptoms - Burning/Stinging

*: Significantly improved from Baseline (p<0.01)
#: Significantly different between groups (p=0.021)

VAS Burning/Stinging scale

15.00  10.00  5.00  0.00  -5.00  -10.00  -15.00
Daytime Ocular Symptoms - Dryness

* : Significantly improved from Baseline (p<0.01)
# : Significantly different between groups (p<0.05)
& : Directionally different between groups (p=0.058)

Figure 10
Figure 11

Lid Wiper Epitheliopathy (LWE)

*: Significantly improved from Baseline (p<0.05)
&: Directionally improved from Baseline (p=0.078)
#: Significantly different between groups (p=0.009)
**INTERNATIONAL SEARCH REPORT**

**PCT/US2014/014505**

**A. CLASSIFICATION OF SUBJECT MATTER**

INV. A61K9/10  A61K47/36  A61K47/38

ADDITIONAL:

According to International Patent Classification (IPC) and both national classification and IPC:

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols):

A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched:

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used):

EPO-Internal, BIOSIS, EMBASE, MEDLINE, WPI Data

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>X</td>
<td>wo 2009/157596 AI (DHP KOREA LTD CO [KR]; SUNG S00 HYUN [KR]; CHOUNG JAI JUN [KR]; KU SAE) 30 December 2009 (2009-12-30)</td>
<td>1-3, 10, 15-20, 22, 23</td>
</tr>
<tr>
<td>Y</td>
<td>example 1 claims 1-15 paragraph [0006] paragraph [0021] - paragraph [0029]</td>
<td>1-29</td>
</tr>
<tr>
<td>Y</td>
<td>example 1 claims 1-102 paragraph [0003]</td>
<td>1-29</td>
</tr>
</tbody>
</table>

Further documents are listed in the continuation of Box C. See patent family annex.

*Special categories of cited documents:

*"A" document defining the general state of the art which is not considered to be of particular relevance.

*"E" earlier application or patent but published on or after the international filing date.

*"L" document which may throw doubts on priority claim(s), or which is cited to establish the publication date of another citation or other special reason (as specified).

*"O" document referring to an oral disclosure, use, exhibition or other means.

*"P" document published prior to the international filing date but later than the priority date claimed.

*"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention.

*"X" document of particular relevance: the claimed invention cannot be considered to be novel or cannot be considered to involve an inventive step when the document is taken alone.

*"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

*"S" document member of the same patent family.

**Date of the actual completion of the international search**

10 March 2014

**Date of mailing of the international search report**

17/03/2014

**Name and mailing address of the ISA**

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016

**Authorized officer**

Sinde, Ulrike
<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>paragraph [0002]</td>
<td>1-29</td>
</tr>
<tr>
<td></td>
<td>paragraph [0018]</td>
<td></td>
</tr>
<tr>
<td></td>
<td>paragraph [0021]</td>
<td></td>
</tr>
<tr>
<td></td>
<td>examples 1,2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>claims 1-13</td>
<td></td>
</tr>
<tr>
<td></td>
<td>examples 10-16</td>
<td></td>
</tr>
<tr>
<td></td>
<td>claims 1-23</td>
<td></td>
</tr>
<tr>
<td>Patent document cited in search report</td>
<td>Publication date</td>
<td>Patent family member(s)</td>
</tr>
<tr>
<td>----------------------------------------</td>
<td>-----------------</td>
<td>-------------------------</td>
</tr>
<tr>
<td>WO 2009157596 A1</td>
<td>30-12-2009</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>BR PI0406636 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CA 2512320 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 1581211 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP 2006516032 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2004137079 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2005260280 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2005266089 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>WO 2004062660 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CA 2687264 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 1992362 A2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 2508205 A2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>ES 2402312 T3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP 2010527363 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2008287672 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>WO 2008144185 A2</td>
</tr>
<tr>
<td>WO 2010047927 A1</td>
<td>29-04-2010</td>
<td>AR 073908 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>AU 2009308017 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CA 2741288 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 2349232 A1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>TW 201021793 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2010184664 A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>WO 2010047927 A1</td>
</tr>
</tbody>
</table>