Title: UV-ABSORBING POLYMERS CONSISTING OF GELATIN AND AMINOBUTADIENE

Abstract: The present invention relates to a process for the preparation of UV-absorbing polymers wherein an aminobutadiene derivative according to formula (I) is reacted with a polymer P having at least one amine group -NHR⁺ according to formula (II). The UV-absorbing polymers are suitable for use in cosmetic formulations, in particular sunscreen formulations.
UV-ABSORBING POLYMERS CONSISTING OF GELATIN AND AMINOBUTADIENΞ

FIELD OF THE INVENTION

The present invention is in the field UV-absorbing compounds, in particular useful in sunscreen compositions.

BACKGROUND OF THE INVENTION

The detrimental effects of exposing the skin to UV light are manifold and are well documented in the prior art. It has been long recognized that UV-B radiation, with a wavelength of 290 to 315 nm, causes erythema or sunburn. It was not until around 1980 that it was discovered that UV-A radiation, with a wavelength from 315 to 400 nm, causes phototoxic and photochemical reactions.

While about 70% UV-B radiation is blocked by the outer skin or stratum corneum, this is not the case for UV-A radiation, which can subsequently penetrate deep into the living dermis. A well known destructive effect of UV-A is oxidative stress. Superoxide, which is formed by UV-A radiation, can release iron from ferritin, an iron-storage protein located in fibroblasts in the skin (Pourzand et al., Proc. Natl. Acad. Sci. USA, June 1999, Vol. 96, p. 6751-56). The role of iron in the Fenton or Haber Weis reaction resulting in the production of highly destructive hydroxyl radicals and hydrogen peroxide is well known. Also other metal ions like copper-ions have been reported to catalyze the formation of oxygen radicals. The role of these destructive products in damaging DNA is well known as for example described by Sestili et al (Free Radical Biology & Medicine [US], July 15 1998, 25, [2] p.196-200).

The normal biochemical protection by enzymes like superoxide dismutase is not sufficient to effectively stop the reaction induced by UV-A radiation. Hence the necessity to protect the skin from these harmful effects is still mandatory. Other oxidative stress phenomena are damaging of collagen resulting for example in accelerated skin aging and white spots, or damaging of cell walls by lipid peroxidation.

Recent interest focuses in particular on 1-amino-1,3-butadienes and derivatives (also known as merocyanine derivatives) thereof. Such compounds are also known in the art, cf. for example US 2,617,827, US 4,045,229, US 4,195,999 and US 4,309,500.

WO 2004/006878 discloses merocyanine derivatives for use in cosmetics for protection against UV radiation, but provides no teachings about the benefits or drawbacks of the numerous listed structures.

However, a disadvantage of many organic UV absorbing compounds is their low water solubility. Therefore, they are unsuitable for use in water-based cosmetics like hydrogels. They require the use of organic solvents, which generally exhibit undesired side effects. Lipid soluble UV absorbing compounds, with or without the combination with organic solvents are capable of passing the so-called stratum corneum with the risk of entering the bloodstream. Consequently, there is a need in the art for more hydrophilic UV-absorbing compounds.

To enhance the hydrophilicity, it is proposed in the art to couple UV absorbing compounds to certain polymeric systems. For example, US 4,839,160 discloses a cosmetic formulation for protecting the skin against UV radiation comprising a polymer of benzylidene norbornanone units having C4 - C12 alkoxy chains. However, this approach does not result in an increase of the hydrophilicity of the UV absorbing compound but into an increase of the hydrophobicity of the UV absorbing compound.

In addition, although the effect on the penetration through the skin is disclosed, the increase of the hydrophobicity will result in an increase of the mobility through the lipid elements in the stratum corneum instead of the other way around.

The coupling of UV-A and UV-B absorbing compounds to a polyacrylic acid through an oxygen or nitrogen atom is disclosed in WO 01/08647, resulting in a molecular complex designed specifically for the uptake of apolar compounds to adapt the refractive index into a desired direction. However, mixing with oily compounds has the risk of skin penetration in case of smaller molecular weight complexes that could result in undesired immunogenic reactions.

WO 03/004061 discloses the use of UV-absorbing complexes wherein a flavonoid is coupled to a polypeptide, in particular a polypeptide selected from the group consisting of casein, sericin, soluble collagen and gelatin. These UV-absorbing complexes have, however, several disadvantages. The coupling method that is disclosed uses formaldehyde as reagent which is a highly toxic material and which
causes strong skin irritations. Furthermore, cross-linking of the polypeptide occurs as an undesired side-reaction. Also, the stability of the complexes is unsatisfactory as has become apparent from stability tests under xenon radiation conditions (see Example 5 of WO 03/004061). Finally, the complexes absorb visible light and this absorption provides the material a yellow colour which is obviously not desired for sunscreen materials in cosmetic applications.

EP A 1172399 discloses a method to couple organic compounds to a polymer which has at least one reactive amine group. In this patent application, the use of the synthesized materials for sunscreen applications is not disclosed or suggested.

US 5,403,944 discloses an organopolysiloxane polymer with UV-absorbing groups.

JP 3161742 discloses UV absorbers combined with gelatin for use in photographic sensitive material. No cosmetic applications are disclosed or suggested.

In spite of these attempts, there remains a need for UV filters which provide sufficient direct protection from sunlight by blocking the radiation while not showing any risk of allergic or immunogenic nature and not showing any penetration through the skin and being fully transparent for the visible light radiation.

WO 2004/075871 discloses a cosmetic composition for protection against UV radiation comprising a cosmetically and dermatologically acceptable carrier and a UV absorbing compound, not being a flavonoid, which is covalently linked to a polypeptide which is preferably selected from the group consisting of casein, sericin, soluble collagen and gelatin. The UV absorbing compound is preferably an aminobutadiene of the formula:

\[
\begin{align*}
\text{R}_1 & \quad \text{N} \\
\text{R}_2 & \quad \text{R}_3 \\
\text{R}_4 & \quad \text{R}_5 \\
\end{align*}
\]

wherein \( \text{R}_1 \) and \( \text{R}_2 \), which may be the same or different, each represents a hydrogen atom, a linear or branched alkyl group having 1 - 20 carbon atoms, or an aryl group having 6 - 20 carbon atoms, provided that \( \text{R}_1 \) and \( \text{R}_2 \) do not simultaneously represent hydrogen atoms, optionally \( \text{R}_1 \) and \( \text{R}_2 \) can combine and form a cyclic amino group; and
wherein each of $R_i$ and $R_2$ may be substituted by one or more carboxy groups (-CO$_2$H) or derivatives thereof;

$R_3$ represents a carboxy group, -COOR$_5$, -COR$_5$ or SO$_2$R$_5$ and $R_4$ represents a carboxy group, -COOR$_5$ or -COR$_5$ wherein $R_5$ and $R_6$, which may be the same or different, each represents a linear or branched alkyl group having 1-20 carbon atoms, or an aryl group having 6 to 20 atoms; optionally $R_5$ and $R_6$ can combine and form a 1,3-dioxocyclohexane nucleus, a barbituric acid nucleus, or a 2,4-diazoo-1-alkoxy-3,5-dioxocyclohexene nucleus. The aminobutadienes are coupled to the polypeptides by a method in which crosslinking of the polypeptide does not occur. In particular, the method involves the conversion of a carboxylic acid containing aminobutadiene to an N-succinimide ester by use of a carbodiimide and N-hydroxy succinimide and subsequent coupling of the ester to the polypeptide.

Non-prepublished WO 2006/009451 (priority date 23 July 2004) discloses UV-absorbing polymers comprising an aminobutadiene moiety and a synthetic amine rich polymer. These UV-absorbing polymers can either be prepared according to the method disclosed in WO 2004/075871 or according to a method wherein the aminobutadiene is directly coupled to the synthetic amine rich polymer having -NHR* end groups, wherein $R^*$ is hydrogen or a linear or branched C$_1$ - C$_6$ alkyl group.

Non-prepublished WO 2006/016806 (priority date 11 August 2004) discloses a particular group of aminobutadiene derivatives for use as UV-absorbing compounds.

**SUMMARY OF THE INVENTION**

The present invention provides an efficacious method for the preparation of UV-absorbing polymers that are based on polymers having at least one amine group -NHR*, wherein $R^*$ is hydrogen or a linear or branched C$_1$ - C$_6$ alkyl group. In particular, the present invention provides a method for the preparation of UV-absorbing polymers that are based on polypeptides.

Accordingly, the present invention provides a process for the preparation of UV-absorbing polymers wherein an aminobutadiene derivative according to formula (I) is
reacted in the presence of a base with a polymer P having at least one amine group -NHR* according to formula (II):

\[
\begin{align*}
\text{(I)} & \quad \text{HR*} - N - P \\
\text{(H)} & \quad \text{R} \quad \text{R} \quad \text{R} \quad \text{R} \quad \text{R} \quad \text{R} \\
\end{align*}
\]

wherein:

R_1 and R_2, which may be the same or different, each represents a hydrogen atom, an optionally substituted linear or branched alkyl group having 1 - 20 carbon atoms, an optionally substituted aryl group having 6 - 20 carbon atoms, or an optionally substituted alkaryl or arylalkyl group having 7 - 20 carbon atoms, provided that R_1 and R_2 do not simultaneously represent hydrogen atoms;

R_3 represents -COOH, -COOR, -COR, CN or SO_2R, and R_4 represents -COOH, -COOR, -COR, CN or SO_2R, wherein R_5 and R_6, which may be the same or different, each represents a linear or branched alkyl group having 1 - 25 carbon atoms, a linear or branched alkyl group having 1 - 25 carbon atoms and one or more carbon-carbon double bonds and/or triple bonds, wherein the alkyl group optionally comprises one or more hetero-atoms selected from the group consisting of O, N, Si and S, an optionally substituted aryl group having 6 to 20 atoms, or an optionally substituted alkaryl or arylalkyl group having 7 - 20 carbon atoms; optionally R_5 and R_6 can combine and form a ring structure, said ring structure preferably being a five to seven membered ring structure, which is optionally substituted and which optionally comprises N, O and/or carbonyl groups;

R* is hydrogen or a linear or branched C_1-C_e alkyl group; and

the polymer P is a polypeptide,

provided that the polymer P is not a synthetic amine rich polymer.

The present invention further relates to the use of the UV-absorbing polymer for the preparation of a cosmetic composition for protection against UV-radiation.
DETAILED DESCRIPTION OF THE INVENTION

The aminobutadiene according to formula (I)

The aminobutadiene according to the present invention has the general formula (I) disclosed above.

If R₁ and R₂ are substituted alkyl groups or substituted aryl groups, the substituents are preferably selected from the group consisting of carboxy (-CO₂H), ester (-CO₂Rs), wherein Rs is a linear or branched C₃ - C₅ alkyl group, a C₆ - C₇ aryl group, a C₇ - C₁₀ alkaryl group or a C₇ - C₁₂ arylalkyl group, or amide (-CO₂N(Rs)₂), wherein the number of substituents is preferably one to three, more preferably one.

A preferred class of aminobutadienes is that in which R₃ represents -COOH, -COOR₅, -COR₅ or SO₂R₅ and R₄ represents a carboxy group, -COOR₆, -COR₆ or SO₂R₆, wherein R₁, R₂, R₅ and R₆ have the meanings indicated above. Examples of aminobutadienes according to the general formula (I) are for example disclosed in US 4,195,999. Table 1 lists a number of aminobutadienes according to the present invention which can be coupled directly to polymer P, including their approximate absorption maxima in methanol (or ethanol when not soluble in methanol). The approximated value is based on measurements for different R₁ and R₂ groups.

Table 1

<table>
<thead>
<tr>
<th>1.1</th>
<th>General structure: R₁ and R₂</th>
<th>R₁ and R₂ used for measuring absorption maximum</th>
<th>Absorption maximum (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.2</td>
<td></td>
<td>n-Hexyl</td>
<td>373</td>
</tr>
<tr>
<td></td>
<td>R1</td>
<td>R2</td>
<td>n-Butyl</td>
</tr>
<tr>
<td>---</td>
<td>-----</td>
<td>-----</td>
<td>---------</td>
</tr>
<tr>
<td>1.3</td>
<td>N-</td>
<td>COOCH₃</td>
<td>SO₂</td>
</tr>
<tr>
<td>1.4</td>
<td>R₁</td>
<td>C₂H₅</td>
<td>R₂</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.5</td>
<td>R₁</td>
<td>C₆H₁₁(t)</td>
<td>R₂</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.6</td>
<td>R₁</td>
<td>N-</td>
<td>R₂</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.7</td>
<td>R₁</td>
<td>N-</td>
<td>R₂</td>
</tr>
<tr>
<td>1.8</td>
<td>R₁</td>
<td>R₂</td>
<td>N-</td>
</tr>
<tr>
<td>1.9</td>
<td>R₁</td>
<td>R₂</td>
<td>N-</td>
</tr>
<tr>
<td>1.10</td>
<td>R₁</td>
<td>R₂</td>
<td>N-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>COOCH₃</td>
</tr>
</tbody>
</table>
Of the aminobutadienes according to the formula (I), an even more preferred class is that represented by the following formula (III):

\[
\begin{align*}
\text{(III)} & \\
\end{align*}
\]

wherein \( R_1, R_2, R_4 \) and \( R_5 \) have the same meaning as in the general formula (I) It was found that the presence of an -SO2R5 group improves the stability of the compounds under UV-light.

A yet even more preferred class of aminobutadienes according to formula (III) is that wherein \( R_4 \) is -COOR5, wherein \( R_6 \) has the meaning identified above.

A yet even more preferred class of aminobutadienes according to the present invention is represented by the general formula (IV), wherein \( R_1, R_2 \) and \( R_3 \) are as defined above and wherein \( R_6 \) is a linear or branched, preferably linear, alkyl group
having 10 - 20 carbon atoms, even more preferably 12 - 20 carbon atoms en most preferably 12 - 18 carbon atoms:

Preferably, at least one of R_i and R_2 is a group of the formula R_7-C(O)-, wherein R_7 is a hydrogen atom or an optionally substituted linear or branched alkyl group having 1 to 20 carbon atoms.

More preferably, one of R_i and R_2 is a group of the formula R_7-C(O)- whereas the other is an optionally substituted phenyl group of 6 to 20 carbon atoms.

A most preferred group of the aminobutadiene according to the formulas (I) and (III) is:

wherein R_6 has the same meaning as in general formula (I). Hereinafter the compound according to formula (V) wherein R_6 is ethyl is referred to as UVI. R_7 can be a hydrogen or a optionally substituted linear or branched alkyl group having 1 to 20 carbon atoms. Another example of the aminobutadiene according to the formulas (I) and (III) is a compound according to formula (V), wherein R_6 is n-CieH_{33}.

If R_7 is a substituted alkyl group, the substituents are preferably selected from the group consisting of carboxy (-CO_2H), ester (-CO_2Rs), wherein R_8 is a linear or branched C_1 - C_5 alkyl group, a C_6 - C_12 aryl group, a C_7 - C_12 alkaryl group or a C_7 -
C12 arylalkyl group, or amide (-CO2N(Rs)2) The number of substituents is preferably one to three, more preferably one.

Preferably, R7 has less than or equal to 12 carbon atoms. More preferably, R7 has 1 to 8 carbon atoms. Even more preferably, R7 has 1, 2, 3, 4 or 5 carbon atoms, because the effect on the absorption maximum is very small when the alkyl group contains more than 6 carbon atoms. Examples of most preferred alkyl groups for R7 are methyl, ethyl, 1-propyl, 2-propyl, n-butyl, s-butyl, t-butyl, 2-methyl-1-propyl, 2-methyl-2-propyl, 1-pentyl, 2-pentyl, 3-pentyl, 2-methyl-1-butyl, 2-methyl-2-butyl, 3-methyl-2-butyl, 3-methyl-1-butyl and 2,2-dimethyl-1-propyl.

The polymer P

According to the present invention, the polymer P is a polypeptide comprising at least one amine group -NHR* as is disclosed above, provided that the polymer P is not a synthetic amine rich polymer as disclosed in non-prepublished WO 2006/009451 (priority date 23 July 2004). Within the context of this invention, it should be understood that the polymer P may not only be of natural origin, but also of synthetic origin, e.g. as can be obtained by recombinant techniques, or a synthetic or natural polymer that is further modified by chemical methods, e.g. grafting techniques.

The molecular weight of polymer P is preferably less than 300 kD, more preferably less than 100 kD, even more preferably less and 50 kD. The molecular weight is preferably at least 1 kD. A more preferred range is a molecular weight of polymer P of 1 to 30, even more preferred is a range of 1 to 20 kD. A molecular weight range of 1.5 to 10 kD is even more preferred. Most preferably, the molecular weight is less than 6 kD, comprising molecular weights of 2.0, 3.0, 4.0 and 5.0 kD.

Within the context of this invention, a polypeptide is to be understood to comprise any molecule or polymer having at least 15 amino acids linked by peptide bonds, including natural proteins, denatured proteins, synthetic and recombinant proteins and peptides, glycoproteins, proteoglycans, lipoproteins and other molecules which may contain other groups, including bio-oligomer or polypeptide groups as a side chain or in the chain. The maximum length is usually 3000 amino acids. Consequently, according to the invention, the polypeptide comprises 10 - 2000 amino acids. Preferably, the polypeptide comprises less than 500 amino acids. Polypeptides
comprising 15 to 100 amino acids are more preferred. Even more preferred are polypeptides comprising less than 60 amino acids.

According to a preferred embodiment of the present invention, the polypeptide is a collagen-like or gelatin-like polypeptide comprising at least one stretch of five or more consecutive repeats of Gly-Xaa-Yaa triplets, wherein it is preferred that in the collagen-like or gelatin-like polypeptide at least 20% of the amino acids are present in the form of consecutive Gly-Xaa-Yaa triplets. Preferably at least 50% of the amino acids are present in the form of consecutive Gly-Xaa-Yaa triplets. A collagen- or gelatin-like polypeptide may have 100% of its amino acids present in the form of consecutive Gly-Xaa-Yaa triplets.

Collagen-like or gelatin-like polypeptides according to the invention are preferably identical or essentially similar to natural human collagen amino acid sequences and non-human sequences (such as rat, rabbit, mouse etc.). Also sequences can be designed that do not occur naturally but which are obtained by recombinant techniques. The term "essentially similar" means that two peptide sequences, when optimally aligned, such as by the programs GAP or BESTFIT using default parameters, share at least 80 percent sequence identity, preferably at least 90 percent sequence identity, more preferably at least 95 percent sequence identity or more (e.g., 99 or 100 percent sequence identity). When calculating the percentage of sequence identity in the context of this invention the threonine residues, serine residues, the residues adjacent to the N-terminal or the C-terminal of threonine and 50% of the residues adjacent to the C-terminal side of the serine residues must be disregarded. Thus for example a natural sequence having a threonine is 100% identical to the same sequence wherein the threonine has been replaced by any other amino acid and the same is true for a sequence having an amino acid-threonine tandem wherein said amino acid is not a glycine compared with a natural sequence having glycine-threonine tandem (assuming the N-terminal is to the left and C-terminal to the right). GAP uses the Needleman and Wunsch global alignment algorithm to align two sequences over their entire length, maximizing the number of matches and minimizes the number of gaps. Generally, the GAP default parameters are used, with a gap creation penalty = 50 (nucleotides) / 8 (proteins) and gap extension penalty = 3 (nucleotides) / 2 (proteins).
A collagen-like or gelatin-like polypeptide in its primary amino acid sequence basically consists of repeats of Gly-Xaa-Yaa triplets, thus approximately one third of the total number of amino acids is a glycine.

Furthermore, characteristic for the collagen-like or gelatin-like polypeptide is the unusual high content of proline residues. Even more characteristic is that in the collagen-like or gelatin-like polypeptide a number of the proline residues are hydroxylated. The most prominent site of hydroxylation is the 4-position resulting in the presence in the collagen-like or gelatin-like polypeptide of the unusual amino acid 4-hydroxyproline. In a triplet 4-hydroxyproline is always found in the Yaa position. Very few proline residues are hydroxylated at the 3 position. In contrast with 4-hydroxyproline, 3-hydroxyproline is always found at the carboxy side of a glycine residue, thus in the Xaa position in a triplet. Different enzymes are responsible for the formation of 3- or 4-hydroxyproline.

Furthermore, another amino acid present in the collagen-like or gelatin-like polypeptide is 5-hydroxylysine. Lysine residues modified in this way are always found in the Yaa position in a triplet.

According to another preferred embodiment of the present invention, the polypeptide is selected from the group consisting of casein, sericin, soluble collagen, gelatin and derivatives thereof. Most preferably, the polypeptide is gelatin or a derivative thereof because of the low antigenicity and immunogenicity, the latter comprising most preferably between 30 and 300 amino acids. If there is a desire to avoid immune reactions even further, for example in case of individuals suffering from (auto-)immune diseases, gelatins incapable of helix-formation can be advantageously used. This can be achieved by chemically modifying gelatins to impair helix formation or, preferably, by using gelatins produced by recombinant techniques as for example disclosed in EP A 1.063.665. Such recombinant gelatins may lack prolines in the peptide backbone or hydroxylation to hydroxy-proline can be reduced or prevented or avoided. Such recombinant gelatins are preferred over modified gelatins because they less likely to elicit immune reactions.

The term "gelatin or a derivative thereof" encompasses materials such as fish gelatin in case the gelling function of the polypeptide is not desired and a larger polypeptide is wished, hydrolysed gelatin, chemically modified gelatin.
Preferably, hydrolysed gelatin has a molecular weight of less than 50 kD and more than 1.5 kD, which is adequate to prevent the skin penetration of the UV absorbing polypeptide complex and showing rather limited or even fully absent gelling properties. Most preferably, hydrolysed gelatin has a molecular weight between 3 kD and 30 kD.

Gelatins that can be used as the polypeptide are preferably acid or lime treated skin or bone gelatins from mammals or cold-blooded animals. Such gelatins are common in the prior art and are well described in literature.

Chemically modified gelatins are also known in the art. Examples include trimellitated, succinylated, acylated, alkylated, phthalated gelatin. Chemically modified gelatins can advantageously be used as long as they do not elicit detrimental reactions like immune reactions when applied to the skin. Succinylated gelatins for example are applied as blood plasma expanders, and are medically and immunologically acceptable gelatin. Alkylated gelatins, cf. for example. DE 19721238 for alkylene succinated gelatin, can be used to alter the interaction of the polypeptide with the skin and hence influence the water fastness of the UV-absorbing polymer. Thus to improve the interaction of the UV-absorbing compounds of the present invention with the skin, alkyl groups, such as for example alkylene succinyl groups, may additionally be coupled to the carrier in the UV-absorbing compound. Alternatives for alkylene succinyl groups are readily known to the skilled person or can easily be identified in terms of having an improved water fastness. Thus in an embodiment the invention relates to UV-absorbing polymers as defined above wherein the polymer P further comprises alkyl groups, preferably alkylene succinyl groups, to improve water fastness, wherein the alkyl groups and the alkylene groups have 4 to 25 carbon atoms.

Modified gelatins in which the amount of free amine-groups has been increased can be used to increase the load of the aminobutadienes according to formula (I) on the gelatin. EP A 487.686 discloses a method to convert carboxy groups into amines. In addition, spacers can be used to increase the amount of free reactive groups in a polypeptide. A spacer is a molecule which can be covalently linked to reactive groups of the polypeptide, like carboxy groups or amine groups, said spacer molecule comprising at least two reactive groups like amine groups or carboxy groups. Examples of suitable spacers are amino acids, e.g. lysine, glutamic acid and aspartic acid.
**Coupling of the aminobutadiene to the polymer P**

According to the invention, the aminobutadiene according to formula (I) is coupled directly to polymer P by mixing the aminobutadiene with the polymer P. The aminobutadiene is coupled to polymer P in an amount of at least about 7.6 mmol per 100 gram polymer P, preferably in an amount of at least about 10 mmol per 100 gram polymer P, more preferably at least about 20 mmol per 100 gram polymer P, even more preferably at least about 40 mmol per 100 gram polymer P and most preferably at least about 100 mmol per 100 gram polymer P. The amount of aminobutadienes used can also be expressed as a percentage of amine groups in polymer P which is bonded to an amino butadiene. Most preferably, 100% of the amine groups is coupled to an aminobutadiene. Generally, good results can already be obtained when more than 50% of the amine groups is linked to an aminobutadiene. The coupled amounts necessary to reach the preferred UV-absorption may obviously vary between different aminobutadiene structures since said aminobutadiene structures can be expected to have varying extinction coefficients.

High molecular weight of polypeptides, i.e. over 100 kD, are less preferred. Such large molecules may give problems as to insufficient penetration in cosmetic applications. The molecular weight of conventional natural or modified polypeptides, in particular gelatins, can be regulated by for example hydrolysis or by enzymatic breakdown of the polypeptide. The desired average molecular weight can be regulated satisfactorily, although the molecular weight distribution can be problematic. After breakdown of the polypeptide too smaller fragments (i.e. lower than about 3 kD), larger fragments can be removed by known techniques such as ultra filtration, dialysis, noodle washing and the like. Recombinant produced gelatins have the additional advantage that the molecular weight and the molecular weight distribution can be regulated precisely. Another method to regulate skin penetration is to adjust the hydrophobicity of the polypeptide by chemical modification of the polypeptide, covalently linking hydrophilic or hydrophobic groups to the polypeptide.

The direct coupling process can be performed in various solvents. Preferred solvents are those in which gelatin and the aminobutadiene dissolve completely. Preferred solvents are those selected from the group consisting of dimethyl formamide...
(DMF), dimethyl acetamide (DMAC), dimethylsulfoxide (DMSO), N-methyl pyrrolidone (NMP), 1,2-ethanediol, tetrahydrofuran (THF), water and mixtures thereof.

The coupling reaction can be carried out in any suitable reaction vessel at a temperature between -20°C and 150°C, depending on the solvent(s) used. At higher temperatures the aminobutadiene may be instable. Preferably, the temperature is between 0°C and 100°C. More preferred are temperatures between 10°C and 80°C. Other suitable temperatures are from 20°C and 30°C. At higher gelatin concentrations temperatures of between than 35°C and 50°C may be preferred.

According to the present invention, a base or a basic compound is present during direct coupling. Use of such basic compounds may increase the coupling yield with at least 30% - 50%. Suitable organic basic compounds comprise tertiary amines such as for example triethylamine and diisopropylethylamine. Suitable inorganic basic compounds are for example carbonate salts and hydroxides.

The preferred amount of basic compound added is between 0.2 and 3 mol equivalents of the aminobutadiene. Preferably, between 0.5 and 2 mol equivalents are added, and most preferably from 0.7 to 1.0 mol equivalents. The amount of organic base necessary to reach a specific yield can be up 1.5 times higher compared to an inorganic base.

Consequently, the method according to the present invention has an advantage over the method disclosed in WO 2004/075871 since no activation steps or other pre-coupling steps are necessary to couple the polymer P to the aminobutadiene.

**The UV-absorbing polymer and cosmetic compositions**

The a UV-absorbing polymer according to the invention that is prepared by coupling an aminobutadiene of formula (I) to the polymer P according to formula (II) has a UV-absorption of at least 5.6 a.u./g.L at 375 nm. This minimum UV-absorption is required to avoid any detrimental effects on the stability and viscosity of the sunscreen formulation. A lower absorption than 5.6 a.u./g.L at 375 nm requires a higher amount of the UV-absorbing polymer in the sunscreen composition giving rise to formulation problems such as stability and viscosity problems. Preferably, the UV-absorbing polymer of this invention has a UV-absorption of at least 20 a.u./g.L, more preferably
of at least 25 a.u./g.L, even more preferably of at least 30 a.u./g.L and most preferably of at least 40 a.u./g.L.

The present invention is also directed to the use of the UV-absorbing polymers according to the invention to protect the human skin or hair from the detrimental effects of exposure to sunlight. The present invention is especially directed to sunscreen compositions comprising stable UV-absorbing polymers that cannot penetrate through the skin into the bloodstream. Preventing the UV absorbing compound from entering the bloodstream reduces the risk of immune reactions or other detrimental effects.

In a preferred embodiment the UV-absorbing polymer absorbs less than 10% of its total absorption above 400 nm. This means that the UV-absorbing polymer is substantially transparent for visible light. In a preferred embodiment, the UV-absorbing polymer has 75% or more of its total absorption in the UV-A region between 315 and 400 nm. Thus the invention relates to a UV-absorbing polymer as defined above wherein less than 10% of the total absorption between 250 and 600 nm of said UV-absorbing compound is above 400 nm. Also the invention relates to a UV-absorbing polymer as defined above wherein at least 75% of the total absorption between 250 and 600 nm of said UV-absorbing compound is between 315 and 400 nm.

The depth of penetration in the skin can be advantageously regulated by controlling the molecular weight of the UV-absorbing polymer which prevents it from entering the bloodstream. In the prior art, no methods are known to control the skin penetration, but allow skin penetration freely. In the present invention even shallow penetration of the skin is prevented.

The UV-absorbing compound can be advantageously used for the preparation of cosmetic or sunscreen compositions to protect the skin or hair from UV radiation.

Various forms of cosmetic compositions for skin protection comprising UV-absorbing compounds are available in the market as lotions, emulsions, creams, milks, gels and the like. These may contain oil and/or alcohol. Also aerosols or sticks are known to be used. All such forms of cosmetic compositions may function as a medium to apply the polypeptide.

One skilled in the art will be able to select suitable cosmetically and dermatologically carriers that can be used in the sunscreen composition of the invention, in particular in combination which polypeptide to which aminobutadiene is linked.
The sunscreen comprising can contain a second, conventional, UV-absorbing compound. This can be a UV-A, UV-B or broadband UV absorbing compound as described in for example EP A 1.055.412. Other additives as applied in the art may also be used.

The cosmetic compositions of the present invention can contain in addition to the UV-absorbing compound various adjuvants conventionally present in cosmetic compositions of this type for example hydrating agents, emollients or thickening agents, surfactants, preservatives, perfumes, dyes, etcetera.

The invention further relates to a sunscreen composition comprising less than 18 wt%, preferably less than 13 wt%, more preferably less than 10 wt% of the UV-polymer compound according to the present invention.

EXAMPLES

Example 1. Direct coupling to gelatin of compound (Ve) [compound (V) in which R₆ is ethyl and R₇ is methyl]

12.5 g of gelatin (hydrolysed Pigskin gelatin, average MW = 2 kD) was dissolved in 47 g of N-methyl pyrrolidone (NMP) at 60°C in 2 hours while stirring. Then, 5 g of compound (Ve) and 1.7 ml of a 25% aqueous solution of sodium carbonate was added. The reaction mixture was stirred for another 2 hours at 50°C. The modified gelatin was precipitated by slowly adding the reaction mixture to 165 ml ethyl acetate. The solid was filtered off and washed two times, by stirring for 20 minutes in 150 ml ethyl acetate. Following the above procedure, 13.0 g UV-gelatin was obtained with a specific absorption E (370 nm, DMSO, 1%/lcm) = 350 a.u.. The specific absorption is determined by UV-spectroscopy at 370 nm and is defined as the absorption of a 1% (10 g/L) solution at a path length of 1 cm.

Example 2. Direct coupling to gelatin of compound (Vi) [compound (V) in which R₆ is isopropyl and R₇ is methyl]

12.5 g of the gelatin (hydrolysed Pigskin gelatin, average MW = 2 kD) was dissolved in 47 g of N-methyl pyrrolidone (NMP) at 60°C in 2 hours while stirring. Then, 5.17 g
of compound (Vi) and 1.7 ml of a 25% aqueous solution of sodium carbonate was added. The reaction mixture was stirred for another 2 hours at 50°C. The modified gelatin was precipitated by slowly adding the reaction mixture to 165 ml ethyl acetate. The solid was filtered off and washed two times, by stirring for 20 minutes in 150 ml ethyl acetate. Following the above procedure 14.5 g UV-gelatin is obtained with a specific absorption $E_{\text{370 nm, DMSO, 1%/1 cm}} = 327$ a.u..

Example 3. Direct coupling to hydrolysed wheat protein of compound (Ve)

12.5 g of the wheat protein (average molecular weight 2 kD, Plantasol W) was dissolved in 47 g of N-methyl pyrrolidone (NMP) at 60°C in 2 hours while stirring. Then, 5 g of compound (Ve) and 1.7 ml of a 25% aqueous solution of sodium carbonate was added. The reaction mixture was stirred for another 2 hours at 50°C. The modified wheat protein was precipitated by slowly adding the reaction mixture to 165 ml ethyl acetate. The solid was filtered off and washed two times, by stirring for 20 minutes in 150 ml ethyl acetate. Following the above procedure 12.0 g UV-protein is obtained with a specific absorption $E_{\text{370 nm, DMSO, 1%/1 cm}} = 160$ a.u..
1. Process for the preparation of UV-absorbing polymers wherein an amino butadiene derivative according to formula (I) is reacted in the presence of a base with a polymer P having at least one amine group -NHR* according to formula (II):

\[
\begin{align*}
R_1 & \quad R_2 \\
\text{N} & \quad R_3 \\
\text{R_4} & \quad \text{HR*N--P}
\end{align*}
\]

wherein:

- \( R_1 \) and \( R_2 \), which may be the same or different, each represents a hydrogen atom, an optionally substituted linear or branched alkyl group having 1 - 20 carbon atoms, an optionally substituted aryl group having 6 - 20 carbon atoms, or an optionally substituted alkaryl or arylalkyl group having 7 - 20 carbon atoms, provided that \( R_1 \) and \( R_2 \) do not simultaneously represent hydrogen atoms;
- \( R_3 \) represents -COOH, -COOR\(_5\), -COR\(_5\), CN or SO\(_2\)R\(_5\) and \( R_4 \) represents -COOH, -COOR\(_5\), -COR\(_5\), CN or SO\(_2\)R\(_5\), wherein \( R_5 \) and \( R_6 \), which may be the same or different, each represents an optionally substituted linear or branched alkyl group having 1 - 25 carbon atoms, an alkyl group having 1 - 25 carbon atoms and one or more carbon-carbon double bonds and/or triple bonds, wherein the alkyl group optionally comprises one or more hetero-atoms selected from the group consisting of O, N, Si and S, an optionally substituted aryl group having 6 to 20 atoms, or an optionally substituted alkaryl or arylalkyl group having 7 - 20 carbon atoms; optionally \( R_5 \) and \( R_6 \) can combine and form a ring structure, said ring structure preferably being a five to seven membered ring structure, which is optionally substituted and which optionally comprises N, O and/or carbonyl groups;
- \( R^* \) is hydrogen or a linear or branched C\(_i\) - C\(_e\) alkyl group; and
the polymer P is a polypeptide, provided that the polymer P is not a synthetic amine rich polymer.

2. Process according to Claim 1, wherein at least one of R\textsubscript{i} and R\textsubscript{2} is a group of the formula R\textsubscript{7}-C(O)-, wherein R\textsubscript{7} is a hydrogen atom or an optionally substituted alkyl group having 1 to 20 carbon atoms.

3. Process according to Claim 1 or Claim 2, wherein one of R\textsubscript{i} and R\textsubscript{2} is a group of the formula R\textsubscript{7}-C(O)- whereas the other is an optionally substituted phenyl group of 6 to 20 carbon atoms.

4. Process according to any one of Claims 1 - 3, wherein the UV-absorbing polymer has a UV-absorption of at least 5.6 a.u./g.L at 375 nm.

5. Process according to any of Claims 1 - 4, wherein the aminobutadiene is represented by the following general formula (III)

\[
\begin{align*}
\text{R}_1 & \text{SO}_2 \\
\text{R}_4 & \text{=C(=N)} \\
\text{N} & \text{R}_2 \\
\end{align*}
\]  

(III)

wherein R\textsubscript{1}, R\textsubscript{2}, R\textsubscript{4} and R\textsubscript{5} are as defined as in claim 1.

6. Process according to any one of Claims 1 - 5, wherein the amino butadiene is represented by the following general formula (IV):

\[
\begin{align*}
\text{R}_1 & \text{SO}_2 \\
\text{R}_6 & \text{OOC} \\
\text{R}_5 & \text{=C(=N)} \\
\text{N} & \text{R}_2 \\
\end{align*}
\]  

(IV)

wherein R\textsubscript{1}, R\textsubscript{2} and R\textsubscript{5} are as defined in Claim 1 and wherein R\textsubscript{6} is a linear or branched alkyl group having 10 - 20 carbon atoms.
7. Process according to any one of Claims 1-6, wherein the aminobutadiene is represented by the following formula (V):

![Chemical Structure Image]

(V)

wherein \( R_6 \) has the same meaning as in Claim 1 and wherein \( R_7 \) is a hydrogen atom or an optionally substituted alkyl group having 1 to 20 carbon atoms.

8. Process according to any one of the preceding claims, wherein the polypeptide is a polymer having at least 15 amino acids up to 2000 amino acids linked by peptide bonds.

9. Process according to Claim 8, wherein the polypeptide is a collagen-like or gelatin-like polypeptide comprising at least one stretch of five or more consecutive repeats of Gly-Xaa-Yaa triplets.

10. Process according to Claim 9, wherein in the collagen-like or gelatin-like polypeptide at least 20% of the amino acids are present in the form of consecutive Gly-Xaa-Yaa triplets.

11. Process according to any one of Claims 1-7, wherein the polypeptide is selected from the group consisting of casein, sericin, soluble collagen, gelatin and derivatives thereof.

12. Process according to Claim 11, wherein the polypeptide is a gelatin or a derivative thereof comprising 30 to 300 amino acids.

13. UV-absorbing polymer obtainable by the process of any one of Claims 1-12.

14. Use of the UV-absorbing polymer according to Claim 13 in cosmetic compositions.

15. Use according to Claim 14, wherein the cosmetic composition is a sunscreen formulation.
16. Cosmetic composition comprising the UV-absorbing polymer according to Claim 13.

17. Cosmetic composition according to Claim 16, wherein the cosmetic composition is a sunscreen formulation.
INTERNATIONAL SEARCH REPORT

PCT/NL2007/050010

A. CLASSIFICATION OF SUBJECT MATTER

INV. C07K14/78

B. FIELDS SEARCHED

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 practical, search terms used)

EPO-Internal, CHEM ABS Data, BIOSIS, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

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Further documents are listed in the continuation of Box C

See patent family annex

* Special categories of cited documents

*A1* document defining the general state of the art which is not considered to be of particular relevance

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*A1* document member of the same patent family

Date of the actual completion of the international search

27 March 2007

Date of mailing of the international search report

04/04/2007

Name and mailing address of the ISA

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Authorized officer

Masturzo, Pietro

Form PCT/ISA/210 (second sheet) (April 2005)
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