

Novel, water-soluble porphyrin platinum compounds with high tumor selectivity and their use for the treatment of benign and malignant tumor diseases

5 Introduction

The invention relates to novel, water-soluble porphyrin platinum compounds with high tumor selectivity and their use for the treatment of benign and malignant tumor diseases. In particular, the inventive compounds are suitable for photodynamic anti-tumor therapy in man and mammals.

Prior art

Platinum(II) complexes with porphyrin ligands and their application as potent cytostatic and phototoxic antitumor agents have already been described in the following publications.

W.M. Sharman, C.M. Allen and J.E. van Lier, DDT 4, (11) 507-517 (1999).
Photodynamic therapeutics: basic principles and clinical applications

T. Okunaka and H. Kato, Rev. Contemp. Pharmacother., 10, 59-68 (1999).
Potential Applications of Photodynamic Therapy.

H. Brunner, H. Obermeier and R.-M. Szeimies, Chem. Ber., 1995, 128, 173-181.

Platinum(II) complexes with porphyrin ligands: synthesis and synergism during photodynamic therapy.

H. Brunner, K.-H. Schellerer and B. Treitinger, Inorg. Chim. Acta 1997, 264, 67-69.

Synthesis and in vitro testing of hematoporphyrin type ligands in platinum(II)

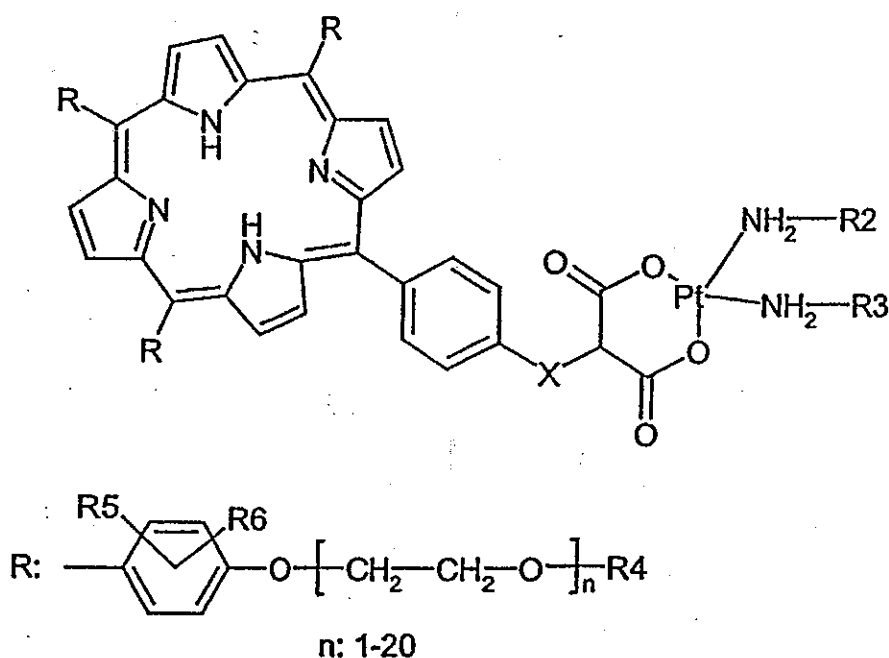
complexes as potent cytostatic and phototoxic antitumor agents.

Description of the invention

In the invention, novel porphyrin platinum derivatives are described, which have cytotoxic properties. Surprisingly, the compounds have good water solubility and a high selectivity. The compounds can be used for the treatment of cancer and, in particular, for the photodynamic treatment of tumors.

The general formulas of the claimed compounds of the tetraarylporphyrin platinum derivatives type are:

10 Formula I



X: O, S, NH, N-Alkyl

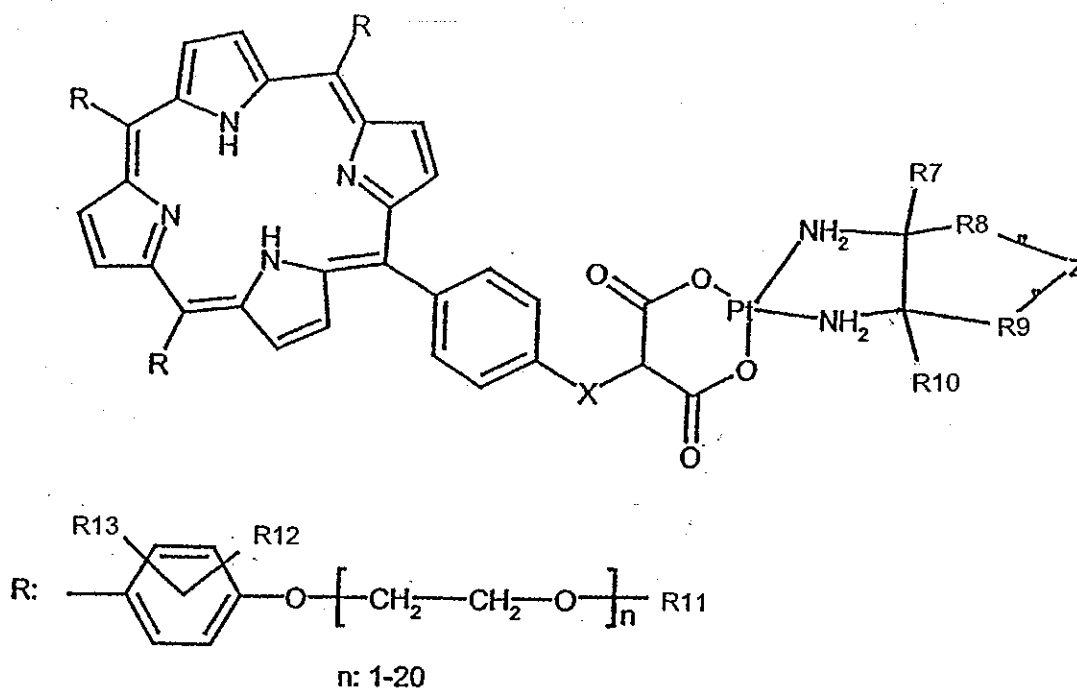
R2/R3: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl

R4: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl

R5: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

R6: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

Formula II



X: O, S, NH, N-Alkyl

R7/R8/R9/R10: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl or R8 and R9 can join together to form a ring with Z = (CH₂)_n and n=0-6 or

R7/R10: H, -(CH₂)_n - COOR14, n=0-6

R11: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl

R12: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

R13: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

R14: H, Alkyl

In a first aspect of the present invention, there is provided porphyrin platinum derivatives of the (a) tetraarylporphyrin platinum derivatives type according to formula I and II as defined above.

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5 If the inventive compounds have at least one center of asymmetry, they can be in the form of their racemates, their pure enantiomers and/or their diastereoisomers or in the form of mixtures of these enantiomers or diastereoisomers.

In a second aspect of the present invention, there is provided use of the compounds as defined above as therapeutic active compounds for the production of medicaments for the treatment of cancer.

In a third aspect of the present invention, there is provided use of the compounds as defined above as therapeutic active compounds for the production of medicaments for the photodynamic treatment of tumors.

10 The inventive compounds exhibit cytotoxic activity in selected tumor cell lines. The antitumor activity is intensified by irradiating with electromagnetic radiation having a wavelength of 600 to 730 nm. The invention accordingly relates to the chemical combination of the cytotoxic principle of the platinum compounds of the cis platinum type with a photodynamically active molecule of the porphyrin derivative type, in such a manner, that compounds of good water solubility and high selectivity are obtained.

The inventive compounds can be administered intraarterially, intracerebrally, intramuscularly, intraperitoneally, intrathecally, intravenously, orally, parenterally, intranasally, rectally, subcutaneously and/or topically in the form of tablets, film-coated tablets, capsules, coated tablets, powders, granulates, drops, syrups, ointments, powders for inhalation, infusion solutions, drinking solutions or in some other suitable form.

In a fourth aspect of the present invention, there is provided medicaments comprising one or more compounds as defined above in addition to customary physiologically tolerable carriers and/or diluents or auxiliaries.

In a fifth aspect of the present invention, there is provided process for the production of the medicament as defined in the fourth aspect is characterized in that one or more compounds as defined above are processed to give pharmaceutical preparations or brought into a therapeutically administrable form using customary pharmaceutical carriers and/or diluents or other auxiliaries.

The synthesis of the inventive compounds is described.

Tetraarylporphyrin platinum derivatives

Synthesis of the substituted benzaldehydes. For the reaction with 4-hydroxybenzaldehyde the respective oligo- and polyethyleneglycol monomethylethers had to be activated at their alcohol terminus with tosyl chloride according to a literature procedure. The etherification was performed by refluxing the tosylated alcohols and 4-hydroxybenzaldehyde together with K_2CO_3 in DMF. The substituted benzaldehydes were separated by filtration and purified by column chromatography.

For platinum coordination to the tetraarylporphyrins to be synthesized it is necessary to introduce two adjacent carboxylic acid groups in one of the substituted benzaldehydes. Therefore, 4-hydroxybenzaldehyde was etherified with diethyl bromomalonate under alkaline conditions. The diethyl 2-(4-

formylphenoxy)malonate was used together with the substituted benzaldehydes for the synthesis of asymmetric tetraarylporphyrins.

Synthesis of the porphyrin ligands. The synthesis of the asymmetric tetraarylporphyrins was performed using the Lindsey method. Pyrrol and the respective benzaldehydes were reacted under Lewis acid catalysis to porphyrinogens, which were oxidized with p-chloranil to the corresponding porphyrins. The tetraarylporphyrin esters were purified by several column chromatographies. The carboxylic acids, which were required for coordination to the platinum(II) fragments, were prepared by hydrolysis of the esters with a mixture of CHCl_3 and 20 % methanolic KOH solution or pure 20 % methanolic KOH solution only.

Synthesis of the platinum fragments. 1,2-Diaminoethane, 1,3-diaminopropane, trans-1,2-diaminocyclohexane and 2,2'-bipyridine were commercially available and used as ligands to prepare the corresponding dichloroplatinum(II) complexes according to literature procedures. Ethyl DL-2,3-diaminopropionate dihydrochloride, ethyl L-2,4-diaminobutanoate dihydrochloride and diethyl meso-4,5-diaminosuberate dihydrochloride were synthesized according to literature procedures and used as ligands for the preparation of the corresponding diiodoplatinum(II) complexes.

Synthesis of the platinum complexes. For the reaction with the porphyrincarboxylic acids cisplatin had to be activated by conversion into diammine(diaqua)platinum(II) hydroxide. It was reacted with an equimolar amount of the porphyrin ligand in a mixture of CHCl_3 , ethanol and water or, in the case of the water-soluble ligand, in pure water. The resulting diammine(malonato)platinum(II) complexes precipitated. To the reaction mixture of the water-soluble complex CH_2Cl_2 was added to remove neutral impurities. The aqueous hphase was evaporated to obtain the product.

The diamine(dichloro)platinum(II) fragments were activated by conversion into diamine(dihydroxy)platinum(II) species, which were reacted with an equimolar amount of the respective porphyrin malonic acid in a mixture of CH_2Cl_2 , ethanol and water or, in the case of the water-soluble ligand, in pure water. The complexes
5 precipitated. To the water-soluble complex CH_2Cl_2 was added to remove neutral impurities, before the aqueous phase was evaporated to obtain the product.

For the reaction with the porphyrinmalonic acids it was necessary to activate the diamine(diiodo)platinum(II) complexes by conversion into diamine(dinitrato)
10 platinum(II) species, which are water-soluble. In this form they were reacted with an equimolar amount of the porphyrin ligands, in a mixture of CH_2Cl_2 , ethanol and water. The water-insoluble complexes precipitated after concentrating the solutions.

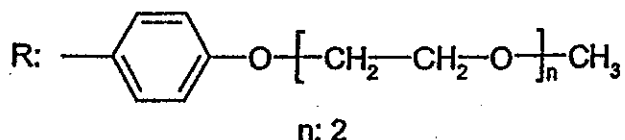
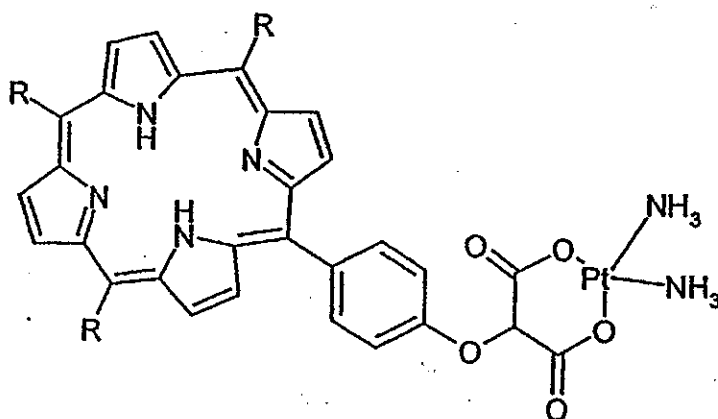
Exemplary embodiments

The following examples are intended to explain the invention in more detail.

The inventive compounds are tetraarylporphyrin platinum derivatives, covered by way of examples 1 and 2.

Examples:

10 Example 1

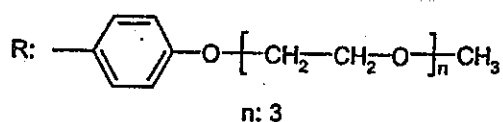
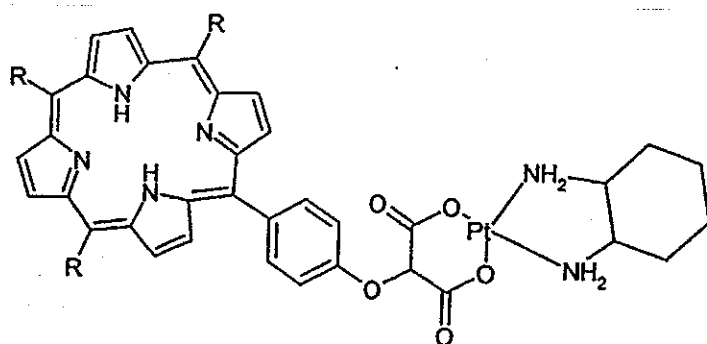


Diammine[2-(4-{10,15,20-tris[4-(1,4,7-trioxaoctyl)phenyl]porphyrin-5-yl}phenoxy)malonato]platinum(II) (No. 21 in Figure 1)

15 The compound 2-(4-{10,15,20-Tris[4-(1,4,7-trioxaoctyl)phenyl]porphyrin-5-yl}phenoxy)malonic acid (109 mg, 0.100 mmol) was dissolved in 10 ml of CHCl_3 and 20 ml of EtOH, combined with 0.100 mmol of the aqueous diammine(diaqua)platinum(II) hydroxide solution and stirred for 20 h. Yield: 81.0 mg (54.2 μmol , 54 %) purple powder, mp 213-214°C.

20 Anal. ($\text{C}_{62}\text{H}_{66}\text{N}_6\text{O}_{14}\text{Pt} \cdot 10 \text{H}_2\text{O}$, 1494.5) C: calcd. 49.83; found. 49.19. H, N: calcd. 5.62; found 6.09.

Example 2



- 5 ($\pm$)-*trans*-1,2-Diaminocyclohexane[2-(4-{10,15,20-tris[4-(1,4,7,10-tetraoxaundecyl)phenyl]porphyrin-5-yl}phenoxy)malonato]platinum(II) (No. 29 in Figure 1).

122 mg (0.100 mmol) of the compound 2-(4-{10,15,20-Tris[4-(1,4,7,10-tetraoxaundecyl)phenyl]porphyrin-5-yl}phenoxy)malonic acid in 10 ml of CH_2Cl_2 and

- 10 20 ml of EtOH were reacted with 0.100 mmol of activated ($\pm$)-*trans*-1,2-diaminocyclohexane(dichloro)platinum(II). Yield: 113 mg (73.9 μmol , 74 %) purple solid, mp 208°C. Anal. ($\text{C}_{74}\text{H}_{86}\text{N}_6\text{O}_{17}\text{Pt}$, 1526.6) C, H, N.

Biological data.

Data of the cytotoxic effect was obtained, for instance, on the human tumor cell lines TCC-SUP and J82. The effect of the compounds was investigated in the dark and
5 under irradiation with light at a wavelength of 600 – 730 nm. Selected compounds are clearly more active cytotoxically under irradiation. There is a synergism between the cytotoxic effect of the platinum component and the photodynamic principle.

Cell lines and general procedures.

- 10 To determine the antiproliferative activity of the new porphyrin ligands and the corresponding platinum complexes with different amine non-leaving groups two bladder cancer cell lines TCC-SUP and J82 were selected as *in vitro* models.

To discriminate between the cytotoxic and phototoxic effects all experiments were
15 carried out in duplicate. The cells were seeded into microplates and the test compounds were added after 48 h. One batch of the microplates was kept in the dark until the end of the experiment, whereas the other microplates were irradiated 48 h after addition of the substances for 10 min with a light dose of $24 \text{ J}\cdot\text{cm}^{-2}$, before the plates were reincubated in the dark.

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End-point chemosensitivity assay.

Tetraarylporphyrin platinum derivatives type.

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At a dosage of 1 μM and 5 μM , both the dark- and phototoxicity of the tetraarylporphyrin-platinum conjugates 21-38 were highly influenced by the type of the non-leaving group the results agreeing with those of the hematoporphyrin-platinum complexes discussed above. 23, 29 and 30 were the most active

30 tetraarylporphyrin-platinum conjugates with $\text{T/C}_{\text{corr.}}$ values of around 37 %, 57 % and

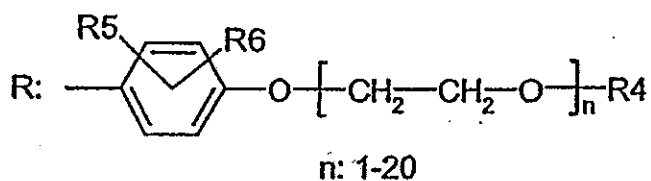
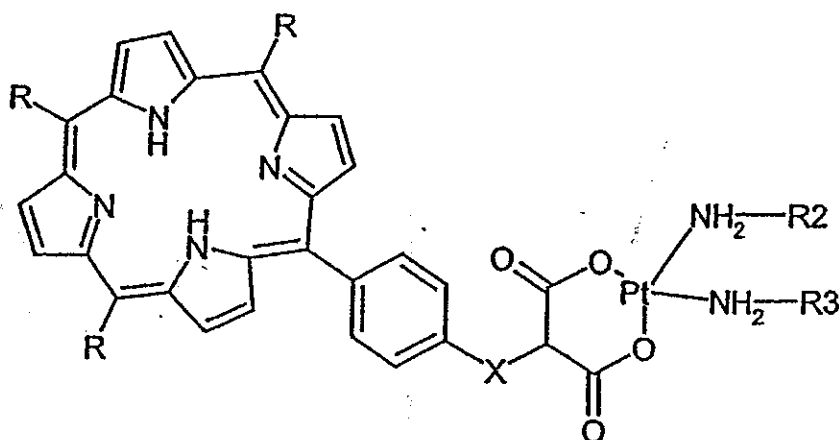
63%, respectively, at 1 μM concentration. At 1 μM concentration there was only a slight enhancement of the cytotoxicity of the tetraarylporphyrin-platinum conjugates with the side chain length $n = 2$ and $n = 3$ upon irradiation. On the average the light-induced T/C_{corr} values were by approximately 20% lower than the dark-only cytotoxicities (data not shown).

An increase in the concentration of the complexes to 5 μM enhanced the dark effects and the phototoxicities as shown in Figure 1. Apart from cisplatin, the highest antitumor activities were measured for the tetraarylporphyrin-platinum conjugates bearing diammine (21-23) and ($\pm$)-trans-1,2-diaminocyclohexane (28-30) non-leaving groups. The differences between the dark and light-induced toxicities were best for the tetraarylporphyrin-platinum complexes 24, 27, 32-34, 36 and 38 with a side chain length of $n = 2$ or $n = 3$ (Figure 1).

Claims

1. Porphyrin platinum derivatives of the

- 5 (a) tetraarylporphyrin platinum derivatives type
according to formula I



X: O, S, NH, N-Alkyl

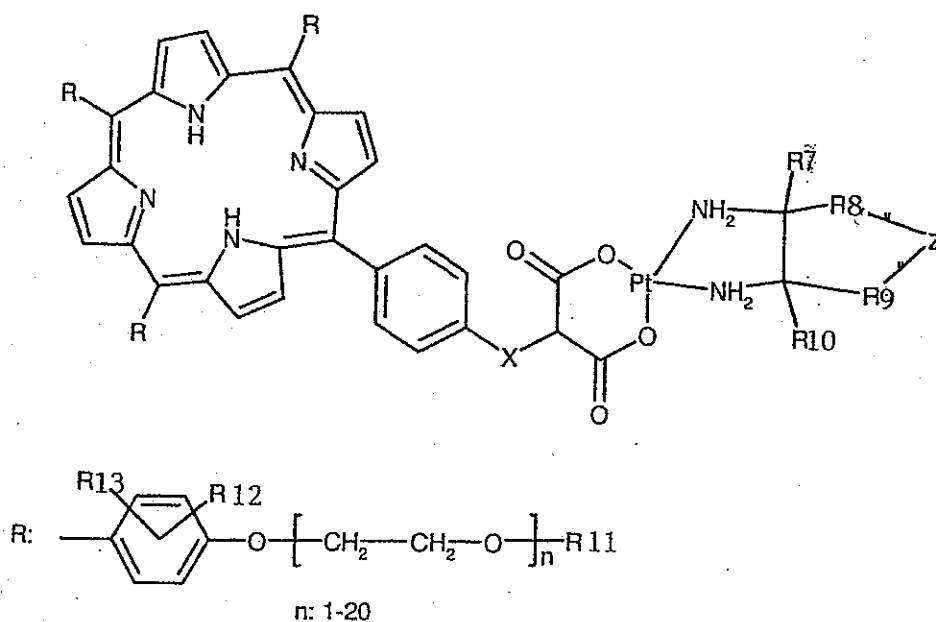
R2/R3: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl

R4: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl

R5: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

R6: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

and according to **formula II**



X: O, S, NH, N-Alkyl

R7/R8/R9/R10: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl or R8 and R9 can join together to form a ring with $Z = (CH_2)_n$ and $n=0-6$ or

R7/R10: H, $-(CH_2)_n - COOR_{14}$, $n=0-6$

R11: H, Alkyl, Aryl, Aralkyl, Hetaryl, Hetarylalkyl, Cycloalkyl

R12: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

R13: H, Alkyl, O-Alkyl, S-Alkyl, Halogen, Nitro, Cyano, Amino, substituted Amino

R14: H, Alkyl

2. Compounds according to claim 1, having one or more asymmetric carbon atoms, in the form of their racemates, their pure enantiomers and/or their diastereoisomers or in the form of mixtures of these enantiomers or diastereoisomers.
3. Use of the compounds according to claims 1 and 2 as therapeutic active compounds for the production of medicaments for the treatment of cancer.

4. Use of the compounds according to claims 1 and 2 as therapeutic active compounds for the production of medicaments for the photodynamic treatment of tumors.
5. Use of the compounds according to claims 1, 2 and 4 as therapeutic active compounds for the production of medicaments for the photodynamic treatment of tumors by irradiating with electromagnetic radiation having a wavelength of 600 to 730 nm.
6. Medicaments comprising one or more compounds according to claims 1 and 2 in addition to customary physiologically tolerable carriers and/or diluents or auxiliaries.
7. Process for the production of a medicament according to claim 6, characterized in that one or more compounds according to claims 1 and 2 are processed to give pharmaceutical preparations or brought into a therapeutically administrable form using a customary pharmaceutical carriers and/or diluents or other auxiliaries.