the combination of targeted therapeutics with light-activated cytosolic delivery induced by photosensitisers preferentially accumulating in solid tumors, PCI can be highly specific and this also contributes to enhanced antitumor efficacy.

As noted above, CCAs are in need of a medical treatment that can provide alternatives to existing methods which, at present, offer limited improvements to life expectancy for patients. The present invention addresses this need.

The present inventors have surprisingly found that, advantageously, a method involving the use of a photosensitizing agent, $TPCS_{2a}$, and gemcitabine at the doses defined herein, and irradiation with light of a wavelength effective to activate the photosensitizing agent results in significant improvements relative to standard treatments. As will be described in more detail in the Examples below, it has been demonstrated that after 6 months of treatment more than 80% of lesions had shrunk and more than 50% of lesions were no longer detectable. This is a remarkable and important result which offers new hope for the treatment of CCAs. The result is particularly surprising as the PCI method relies on release of molecules in the endosome into the cytosol and there was nothing to suggest that gemcitabine was taken up in cells into the endosome and hence could benefit from PCI treatment.

Thus, disclosed is a method of treating a cholangiocarcinoma in a human patient comprising:
i) systemically administering $TPCS_{2a}$ to said patient to a dose of 0.05 to 0.5 mg/kg (or 0.1 to 0.5 mg/kg) and
ii) after 3-5 days, systemically administering gemcitabine to a dose of 500-1500 mg/m^2 and irradiating said cholangiocarcinoma with light with a wavelength of 640-665nm using an optical fiber placed within 3cm of said cholangiocarcinoma to provide a light dose of 10 to 60 J/cm (or 15 to 60 J/cm) (e.g. cm of the fiber or tumour); and optionally
iii) after 1-40 days (preferably after 7-21 days) systemically administering gemcitabine and/or another cytotoxic agent, preferably cisplatin.

In one aspect, there is provided use of gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent, preferably cisplatin, in the manufacture of a medicament for treating a cholangiocarcinoma in a human patient, wherein
i) said $TPCS_{2a}$ is to be systemically administered to said patient to a dose of 0.05 to 0.5 mg/kg; and
ii) after 3-5 days, said gemcitabine is to be systemically administered to said patient to a dose of 500-1500 mg/m^2 and said cholangiocarcinoma is to be irradiated with light with a wavelength of 640-665nm using an optical fiber placed within 3cm of said cholangiocarcinoma to provide a light dose of 10 to 60 J/cm; and optionally
iii) after 1-40 days (preferably after 7-21 days) gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be systemically administered to said patient.

As defined herein "treating" refers to reducing, alleviating or eliminating one or more symptoms of the CCA which is being treated, relative to the symptoms prior to

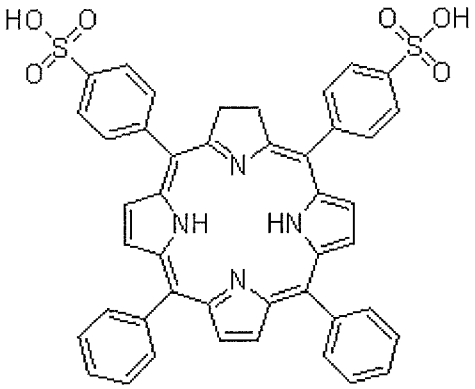
treatment. In particular said treatment may comprise reduction in the size or volume of the CCA being treated. The treatment may include effects on the CCA closest to the optical fiber as well as other CCAs which are more distant and may not directly receive irradiation from the light source. In one embodiment of the disclosure the method treats one or more cholangiocarcinoma (or cells therefore) which are not irradiated during the irradiation step (this includes direct or indirect irradiation, i.e. which do not receive a light dose resulting from the irradiation step).

A "cholangiocarcinoma" is a bile duct cancer which may be intrahepatic or extrahepatic (which may be perihilar and distal). Over 90% of CCAs are adenocarcinomas.

The "patient" is a human.

"Systemic administration" includes any form of non-local administration in which the agent is administered to the body at a site other than directly adjacent to, or in the local vicinity of, the CCA, resulting in the whole body receiving the administered agent. Conveniently, systemic administration may be via enteral delivery (e.g. oral) or parenteral delivery (e.g. intravenous, intramuscular or subcutaneous). Intravenous delivery is preferred.

"$TPCS_{2a}$" is a photosensitizing agent (tetraphenyl chlorin disulphonic acid) having the structure below or isomers thereof. The structure below provides the 7,8-dihydro isomer. The 12,13- and 17,18-dihydro isomers are encompassed by the term $TPCS_{2a}$. Pharmaceutically acceptable salts thereof are also encompassed. Such molecules are as described in WO03/020309 and WO2011/018635 which are incorporated herein by reference.



$TPCS_{2a}$ may be obtained from BOC Sciences, NY, USA or from PCI Biotech AS, Norway. Alternatively, $TPCS_{2a}$ may be prepared as described in WO03/020309 and WO2011/018635 . Pharmaceutically acceptable salts include acid addition salts with physiologically acceptable organic or inorganic acids. Suitable acids include, for example, hydrochloric, hydrobromic, sulphuric, phosphoric, acetic, lactic, citric, tartaric, succinic, maleic, fumaric and ascorbic acids. Hydrophobic salt may also conveniently be produced by, for example, precipitation. Appropriate salts include, for example, acetate, bromide, chloride, citrate, hydrochloride, maleate, mesylate, nitrate, phosphate, sulphate, tartrate, oleate, stearate, tosylate, calcium, meglumine, potassium and sodium salts. Procedures for salt formation are conventional in the art.

Preferred salts include diethanolamine salt, ethanolamine salt (preferably bis (monoethanolamine)), N-methyl-glucamine salt, triethanolamine salt, 1-(2-hydroxymethyl)-pyrrolidine salt and 2-amino-2-(hydroxymethyl) propane-1,3-diol salt.

"Pharmaceutically acceptable" as referred to herein refers to ingredients that are compatible with other ingredients used in the methods or uses of the disclosure as well as physiologically acceptable to the recipient.

The agents used herein, e.g. the $TPCS_{2a}$, gemcitabine and/or other cytotoxic agent may be provided in the form of a pharmaceutical composition for use in the method of the disclosure and may be formulated in any convenient manner according to techniques and procedures known in the pharmaceutical art, e.g. using one or more pharmaceutically acceptable diluents, carriers or excipients. "Pharmaceutically

Although not wishing to be bound by theory, it is likely that gemcitabine is taken up into intracellular compartments in the patient's cells and activation of the photosensitizing agent releases the gemcitabine into the cell. As a consequence, before irradiation is conducted the photosensitizing agent must be in the relevant compartments in the cell. It has been found that molecules in those compartments at the time of activation of the photosensitizing agent, or which are taken up into such cells immediately after such activation, are released into the cytosol. (See in this regard WO02/44396, which is incorporated herein by reference which refers to the so-called "light before" method.)

Conveniently said irradiation is conducted within the time range of 1 hour before the start of gemcitabine administration up to 4, 5 or 6 hours after the start of gemcitabine administration. Preferably the irradiation is performed within 3 or 4 hours of the start of gemcitabine administration (i.e. from the start of gemcitabine administration to 3 or 4 hours after the start of gemcitabine administration, e.g. at time 0, 30, 60, 120, 180 or 240 minutes when time 0 is the start of gemcitabine administration). Thus, for example, when gemcitabine administration is conducted for 30 minutes, and irradiation is conducted for 15 minutes, the gemcitabine administration may be started at time 0, stopped at time 30 mins, irradiation started at time 120 minutes and concluded at time 135 minutes. However, irradiation performed up to 1, 2, 3 or 4 days after gemcitabine administration is also encompassed by the method of the disclosure.

Where appropriate the light irradiation step may be performed more than once (e.g. 2, 3 or 4 times). This may be necessary for particularly large tumours or discrete tumours spread over a wide area. In this case one or more optical fibers may be used which may be at the same or different locations. The location of the one or more optical fibers may be changed or maintained for each irradiation step (e.g. moved to a location close to other discrete tumours or areas of a single tumour mass).

The light irradiation step to activate the photosensitising agent may take place according to techniques and procedures well known in the art. The wavelength of light to be used is 640-665 nm, preferably 652 nm. Suitable artificial light sources

are well known in the art. Conveniently, illumination (irradiation) is provided by the PCI Biotech AS 652nm laser system diode laser, although any suitable red light source may be used.

An optical fiber is used to provide the light which is placed within 3 cm of the CCA. As referred to herein an "optical fiber" is a thin, flexible, transparent fiber through which light can be transmitted at the distal end. Preferably the light is diffused to spread light evenly across a surface, minimizing or removing high intensity bright spots. Various tips at the end of the optical fiber may be used, e.g. a frontal distributor or microlense tip or a spherical diffuser. In the alternative a balloon catheter may be used to scatter the light (e.g. Medlight's diffusing balloon catheter). In another alternative the fiber may be unadorned, e.g. a bare tip may be used. Preferably the light is transmitted through a cylindrical diffuser in which the distal extremity of the fiber is an illumination tip which uniformly distributes the transmitted light along its length.

The fiber may be made of glass or plastic and has a core diameter which is at least the diameter of the laser output channel, preferably of 200 to 800 μm (e.g. 400-600, e.g. 500 μm) or a diameter of 400 to 1200 μm (e.g. 900 to 1000 μm) including any coating. Fibers with diffuser lengths of 10 to 70 mm, preferably 20 to 40 mm may be used. More than one optical fiber may be used for large tumours or multiple discrete tumours within a patient, e.g. 2 or more, e.g. 3, 4, 5, 6 or more, e.g. less than 10. In the alternative a single optical fiber may be used which may be moved to different locations, or maintained at the same location, for multiple rounds of illumination (irradiation).

In accordance with the disclosure the fiber (or at least one or each fiber) is placed within 3 cm of the CCA. This measurement refers to the distance between the exterior surface of the fiber and the closest portion of the CCA. Preferably the fiber is placed as close as possible to the CCA, e.g. within 1 or 2 cm of the CCA or within the CCA. Conveniently the fiber is placed in the bile duct (e.g. via a catheter).

The optical fiber may be provided by way of a catheter, e.g. in the form of an optical fiber based catheter which may be coupled with a laser source (i.e. such that the optical fiber is within the catheter during treatment and light irradiation from the fiber

occurs through the catheter wall). The optical fiber guides the light from the proximal to the distal end of the device. The distal end preferably has an illumination tip ("diffuser") which may uniformly distribute light transmitted by the optical fiber along its length. The catheter allows endoscopic delivery of light. Preferably a light diffuser is used. Conveniently, Medlight SA (Switzerland) Cylindrical Light Diffusers may be used, e.g. the Cylindrical light diffuser Model RD with radiopaque markers.

The time for which the cells of the patient are exposed to light in the methods of the present disclosure may vary to achieve the required light dose. The time may be selected according to various factors including the dose of the photosensitizing agent and gemcitabine and fluence rate of the light to be used and the proximity of the optical fiber to the tumour.

The total light dose to the tumour cells may be expressed in J/cm of the fiber (or tumour) and is calculated as the fluence rate (W/cm of the fiber or tumour) x treatment time.

Depending on the fluence rate of the light source to be used, to achieve the desired light dose, the time of irradiation may be selected accordingly. For example, to achieve a light dose of 10 or 30 J/cm (or 15 or 30 J/cm) with a fluence rate of 100 mW/cm, an irradiation time of 2.5 minutes or 5 minutes, respectively, may be used. Thus, with a fluence rate of 100 mW/cm to achieve a light dose of 10 to 60 J/cm (or 15 to 60 J/cm) an irradiation time of 2.5 to 10 minutes may be used. A higher or lower fluence rate allows a lower or higher irradiation time to be used, respectively. Preferably, taking the above into account, the irradiation time is from 1 minute to 20 minutes, e.g. 2 to 10 minutes, depending on the fluence rate of the light source. These timings refer to each irradiation time when multiple rounds of illumination (irradiation) are used.

Appropriate light doses can be selected by a person skilled in the art and again will depend on the factors indicated above. In particular higher doses or concentrations of the photosensitizing agent ($TPCS_{2a}$) allow a lower light dose to be used. In the present disclosure it has been found that light at a dose of 10 to 60J per cm (or 15 to 60J per cm) of the (diffuser of the) optical fiber or tumour is particularly

advantageous. In the unit "J per cm" the cm is the length of the optical fiber if light is emitted over a length of the fiber (e.g. when a diffuser is used). In the alternative this may be cm of tumour, e.g. when a point light source is used. In both cases J/cm refers to the light dose provided per cm to the local environment. Conveniently the light dose is achieved using a light diffuser with a fluence of 100mW per cm of fiber (e.g. diffuser) and hence an irradiation time of from 2.5 to 10 minutes. Preferably a dose of 10 to 45 (or 15 to 45), e.g. 20 to 40 or 25 to 35 J/cm is used.

Furthermore, if cell viability is to be maintained, the generation of excessive levels of toxic species is to be avoided and the relevant parameters may be adjusted accordingly.

The methods of the disclosure may inevitably give rise to some cell damage by virtue of the photochemical treatment i.e. by photodynamic therapy effects through the generation of toxic species on activation of the photosensitizing agent. As the role of the method of the disclosure is to destroy tumour cells, this cell death may not be of consequence and may indeed be advantageous. In some embodiments, however, cell death should be avoided to ensure that the cytotoxic molecules are taken up into the cells to ensure localized and specific cell death. The methods of the disclosure may be modified such that the fraction or proportion of the surviving cells is regulated by selecting the light dose in relation to the dose (concentration) of the photosensitizing agent. Again, such techniques are known in the art.

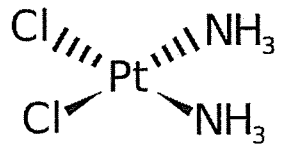
Preferably, substantially all of the cells, or a significant majority (e.g. at least 75%, more preferably at least 80, 85, 90 or 95% of the cells) are not killed by the photochemical internalization method alone (i.e. without a cytotoxic agent). *In vitro* cell viability following PCI treatment can be measured by standard techniques known in the art such as the MTS test. *In vivo* cell death of one or more cell types may be assessed within a 1cm radius of the point of administration (or at a certain depth of tissue), e.g. by microscopy or other appropriate means. As cell death may not occur instantly, the % cell death refers to the percent of cells which remain viable within a few hours of irradiation (e.g. up to 4 hours after irradiation) but preferably refers to the % viable cells 4 or more hours after irradiation.

In optional methods of the disclosure, 1-40 days after step (ii), i.e. after administering the gemcitabine and subjecting the patient to irradiation, the patient may be systemically administered gemcitabine and/or another cytotoxic agent. Preferably such administration occurs 5-30 days, especially preferably 7-21 days after step (ii).

When gemcitabine is administered this may be administered as described hereinbefore for the gemcitabine administered in step ii) (i.e. as regards the dose, route and duration of administration).

The other cytotoxic agent may be any toxic agent which is suitable for treating a cancer, particularly cholangiocarcinoma with acceptable side-effects in the patient (e.g. commonly used chemotherapy drugs). Such agents include alkylating drugs, anthracyclines and other cytotoxic antibiotics (e.g. bleomycin), a protein toxin (e.g. gelonin), antimetabolites (not including gemcitabine), vinca alkaloids and etoposide, tyrosine kinase inhibitors and other antineoplastic drugs such as platinum compounds, e.g. cisplatin.

"Cisplatin" is a platinum containing anti-cancer drug with the structure indicated below ((*SP*-4-2)-diamminedichloroplatinum(II)).



Cisplatin is available from sources such as Hospira (Cisplatin Hospira). Appropriate doses for the other cytotoxic agents are known in the art. If cisplatin is used, conveniently it is used at a dose of 10-50 mg/m^2, preferably 20-30 mg/m^2. The route and duration of administration may be as described hereinbefore for gemcitabine.

In step iii) when gemcitabine and at least one other cytotoxic agent is used, they may be used simultaneously, separately or sequentially. When used simultaneously they are administered at the same time, but may be administered by a single route or via separate routes (e.g. a mixture administered intravenously or

two (or more) preparations administration at the same time but via different intravenous entry points). When administered separately they may be administered at the same time or sequentially and/or may overlap in their administration timing. When used sequentially the time separating administration of the different agents is not more than 24 hours. Conveniently they are administered together in a single mixture.

In preferred features of the disclosure the gemcitabine and/or another cytotoxic agent used in step iii) is administered more than once, e.g. 2, 3, 4, 5, 6, 7, 8, 9 or 10 times (e.g. up to 20 times). This administration may be in a single (or each) cycle or in total in multiple cycles.

As referred to herein a "cycle" is a time period over which a particular treatment regime is applied and is generally repeated to provide cyclical treatment. The treatment in each cycle may be the same or different (e.g. different dosages, timings etc. may be used). A cycle may be from 14-30 days in length, e.g. a 21 day cycle. Multiple cycles may be used, e.g. at least 2, 3, 4 or 5 cycles, e.g. 6, 7, 8, 9 or 10 (e.g. up to 8, 9, 10 or 20) cycles. Within each cycle the gemcitabine and/or another cytotoxic agent may be administered once or more than once, as described hereinbefore. In a preferred feature the gemcitabine and/or another cytotoxic agent is administered twice in each cycle and preferably at last 3, preferably at least 5 cycles are conducted.

Whilst in a preferred feature the above step iii) treatment with gemcitabine and/or another cytotoxic agent is performed, in another embodiment of the disclosure such treatment is not performed and in particular the treatment does not include the use of another cytotoxic agent such as cisplatin.

For effective treatment, the method described herein, or a part thereof, may be repeated. Thus, for example, steps i) and ii) may be repeated one or more times, e.g. 2 or 3 times, for example after an interval of from 1 to 6 months or longer, e.g. after step ii) or iii). In a preferred embodiment the entire method is repeated at least twice, e.g. 3 or 4 times.

In a particularly preferred embodiment the method comprises performance of the method twice (i.e. performance of steps i), ii), iii), i), ii) and iii)) or more, preferably in which in the first and/or second (or later) round of steps i) to iii), step iii) is performed at least twice. Thus by way of example, the method may be performed as follows:

i) systemically administering $TPCS_{2a}$ to said patient to a dose of 0.05 to 0.5 mg/kg (or 0.1 to 0.5 mg/kg) and

ii) after 3-5 days, systemically administering gemcitabine to a dose of 500-1500 mg/m^2 and 2-4 hours after completion of the gemcitabine administration irradiating said cholangiocarcinoma with light with a wavelength of 640-665nm using an optical fiber placed within 3cm of said cholangiocarcinoma to provide a light dose of 10 to 60 J/cm (or 15 to 60 J/cm);

iii) after 1-40 days (preferably after 7-21 days) systemically administering gemcitabine and/or another cytotoxic agent, preferably cisplatin;

iv) repeating step iii) at least once; and

v) repeating steps i) to iv) at least once.

In a particularly preferred embodiment, step iii) is performed in cycles as described hereinbefore, in particular multiple cycles of step iii) are performed (before commencing to step iv)) and in each of these cycles the gemcitabine and/or another cytotoxic agent such as cisplatin is administered twice or more.

By way of example the method may be performed in which step iii) in the first and second round comprises 2, 3 or 4 cycles of gemcitabine and cisplatin administered once or twice, e.g. at days 1 and 8. When steps i) and ii) are performed after the first round step iii) and before the second round step iii), it may impinge on the first cycle in step iii) of the second round, e.g. the gemcitabine administration and irradiation may replace the first administration of gemcitabine and cisplatin in the first cycle. In one option, the patient is not subjected to any treatment for a period of between 1 week to 4 weeks after the first round (of steps i), ii) and iii)) before the second round is commenced. In that case the treatment then resumes with steps i) and ii) as indicated above, followed by step iii). Preferred timings of the treatment and/or doses to be used are as described in the Examples.

Prior to treatment the patient to be treated may be subjected to biliary stenting to ensure adequate biliary drainage. The stent, which may be plastic or metal, is

placed into the bile duct using procedures known in the art. The latter may be kept in place during illumination whereas the former are removed during illumination and a new stent inserted after that procedure.

The above methods and preferred features thereof also apply to uses of the above described agents. Thus, in an alternative aspect the present invention provides gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent, preferably cisplatin, for use in treating a cholangiocarcinoma in a human patient, wherein i) said $TPCS_{2a}$ is to be systemically administered to said patient to a dose of 0.05 to 0.5 mg/kg (or 0.1 to 0.5 mg/kg); and
ii) after 3-5 days, said gemcitabine is to be systemically administered to said patient to a dose of 500-1500 mg/m^2 and said cholangiocarcinoma is to be irradiated with light with a wavelength of 640-665nm using an optical fiber placed within 3cm of said cholangiocarcinoma to provide a light dose of 10 to 60 J/cm (or 15 to 60 J/cm) (e.g. of the fiber or tumour); and optionally
iii) after 1-40 days (preferably after 7-21 days) gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be systemically administered to said patient.

The above described definitions and preferred features are similarly applicable.

Disclosed is a kit comprising gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent as defined hereinbefore, preferably for simultaneous, separate or sequential use to treat a cholangiocarcinoma in a patient, wherein preferably said use is as defined hereinbefore.

In another aspect, there is provided a kit comprising gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent as described herein, preferably for simultaneous, separate or sequential use in treating a cholangiocarcinoma in a patient, wherein preferably said use is as described herein.

Preferred embodiments according to the disclosure are as set out in the Examples in which one or more of the parameters or components used in the Examples may be used as preferred features of the methods described hereinbefore.

The invention will now be described in more detail in the following non-limiting Examples with reference to the following drawings in which:

Figure 1 shows cytotoxicity after 72 hours gemcitabine exposure. Viability of the cell lines TFK-1 (A) and EGI-1 (B) were analysed by the MTT assay after 72 hours gemcitabine incubation as described under Materials and Methods in Example 1.

Claims:

1. Use of gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent, preferably cisplatin, in the manufacture of a medicament for treating a cholangiocarcinoma in a human patient, wherein
i) said $TPCS_{2a}$ is to be systemically administered to said patient to a dose of 0.05 to 0.5 mg/kg; and
ii) after 3-5 days, said gemcitabine is to be systemically administered to said patient to a dose of 500-1500 mg/m^2 and said cholangiocarcinoma is to be irradiated with light with a wavelength of 640-665nm using an optical fiber placed within 3cm of said cholangiocarcinoma to provide a light dose of 10 to 60 J/cm; and optionally
iii) after 1-40 days (preferably after 7-21 days) gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be systemically administered to said patient.

2. Gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent, preferably cisplatin, for use in treating a cholangiocarcinoma in a human patient, wherein i) said $TPCS_{2a}$ is to be systemically administered to said patient to a dose of 0.05 to 0.5 mg/kg; and
ii) after 3-5 days, said gemcitabine is to be systemically administered to said patient to a dose of 500-1500 mg/m^2 and said cholangiocarcinoma is to be irradiated with light with a wavelength of 640-665nm using an optical fiber placed within 3cm of said cholangiocarcinoma to provide a light dose of 10 to 60 J/cm; and optionally
iii) after 1-40 days (preferably after 7-21 days) gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be systemically administered to said patient.

3. The use of claim 1, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in claim 2, wherein said $TPCS_{2a}$ is to be administered to a dose of 0.05 to 0.3 mg/kg, preferably 0.2 to 0.3 mg/kg.

4. The use of claim 1 or 3, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in claim 2 or 3, wherein said gemcitabine in step ii) is to be administered to a dose of 900-1100 mg/m^2.

5. The use of any one of claims 1, 3 and 4, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to

4, wherein said irradiation is to be performed within 4 hours of the start of gemcitabine administration.

6. The use of any one of claims 1 and 3 to 5, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 5, wherein the wavelength of said light is 652nm.

7. The use of any one of claims 1 and 3 to 6, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 6, wherein said light dose is 10 to 45, preferably 25 to 35 J/cm.

8. The use of any one of claims 1 and 3 to 7, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 7, wherein in step iii) both gemcitabine and another cytotoxic agent, preferably cisplatin, are to be administered.

9. The use of any one of claims 1 and 3 to 8, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 8, wherein in step iii) the gemcitabine is to be administered to a dose of 500-1500 mg/m^2, preferably 900-1100 mg/m^2.

10. The use of any one of claims 1 and 3 to 9, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 9, wherein in step iii) the cytotoxic agent is cisplatin which is to be administered to a dose of 10-50 mg/m^2, preferably 20-30 mg/m^2.

11. The use of any one of claims 1 and 3 to 10, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 10, wherein in step iii) said gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be administered more than once.

12. The use, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in claim 11, wherein said gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be administered more than once (preferably twice) in a 14-30 day cycle that follows 1-40 days after steps i) and ii).

13. The use, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in claim 10 or 11, wherein said gemcitabine and/or another cytotoxic agent, preferably cisplatin, is to be administered once or more (preferably twice) in at least 3, preferably at least 5, 14-30 day cycles, wherein said first 14-30 day cycle follows 1-40 days after steps i) and ii).

14. The use of any one of claims 1 and 3 to 13, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 13, wherein steps (i), (ii) and (iii) are to be performed at least twice, wherein preferably in each round of steps (i), (iii) and (iii), step (iii) is to be performed at least twice.

15. The use of any one of claims 1 and 3 to 14, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2-14, wherein the systemic administration of one or more of $TPCS_{2a}$, gemcitabine and another cytotoxic agent, preferably cisplatin, is to be intravenous.

16. The use of any one of claims 1, 3 to 9 and 11 to 15, or the gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent for use as claimed in any one of claims 2 to 9 and 11 to 15, wherein cisplatin is not to be used in said treatment.

17. A kit comprising gemcitabine and $TPCS_{2a}$ and optionally another cytotoxic agent as defined in any one of claims 1-4, 9 and 10, preferably for simultaneous, separate or sequential use in treating a cholangiocarcinoma in a patient, wherein preferably said use is as defined in any one of claims 1 to 16.