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Short communication

Synthesis and biological evaluations of a monomethylauristatin E glucuronide prodrug for selective cancer chemotherapy

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ABSTRACT

We developed a glucuronide prodrug of the potent monomethylauristatin E (MMAE). This prodrug is significantly less toxic than the parent drug. However, in the presence of β-glucuronidase the prodrug leads to the efficient release of MMAE thereby triggering a subnanomolar cytotoxic activity against several cancer cell lines. Preliminary *in vivo* experiments conducted in C57BL/6 mice bearing a subcutaneous murine Lewis Lung Carcinoma (LLC) demonstrated the potential of this targeting system for the selective treatment of solid tumors.

1. Introduction

The development of new anticancer agents allowing the destruction of tumor cells without affecting normal tissues represents one of the main challenges of cancer chemotherapy. In this context, the design of nontoxic drug carriers programmed for both the recognition of a malignant specificity and the release a potent anti-neoplastic agent exclusively in the tumor area has attracted considerable attention over the last decades [1]. The efficiency of such a targeting strategy was recently demonstrated in human with an antibody-drug conjugate [2], Brentuximab Vedotin [3] which reached the market in 2011 for the treatment of lymphomas.

The vast majority of the targeting systems that have been studied so far were designed to recognize cancer cell surface specificities such as tumor-associated membrane receptors or antigens. However, since cancerous tissues are highly heterogeneous, the sole destruction of a particular population of malignant cells could limit the efficiency of this therapeutic approach. Another promising strategy relies on the use of nontoxic prodrugs that can be selectively activated by an enzyme naturally overexpressed in the tumor microenvironment. In this case, the parent drug is released in the extracellular medium and can further penetrate passively inside a wide diversity of surrounding malignant cells whatever their membrane characteristics. Within this framework, β-glucuronidase located in high concentration in the microenvironment of numerous solid tumors including lung, breast, ovarian and gastrointestinal tract carcinomas, is an attractive target to develop enzyme-responsive drug carriers [4]. The potential of this approach [5] was already demonstrated in mice with several glucuronide prodrugs which led to superior therapeutic efficacy compared to standard treatment [6]. However, the poor turnover of β-glucuronidase in the tumor microenvironment represents the "Achilles' heel" of this targeting strategy. Above a certain threshold dose, the enzyme is saturated, therefore limiting the β-glucuronidase-mediated conversion of the prodrug into the parent cytotoxic. As a result, the

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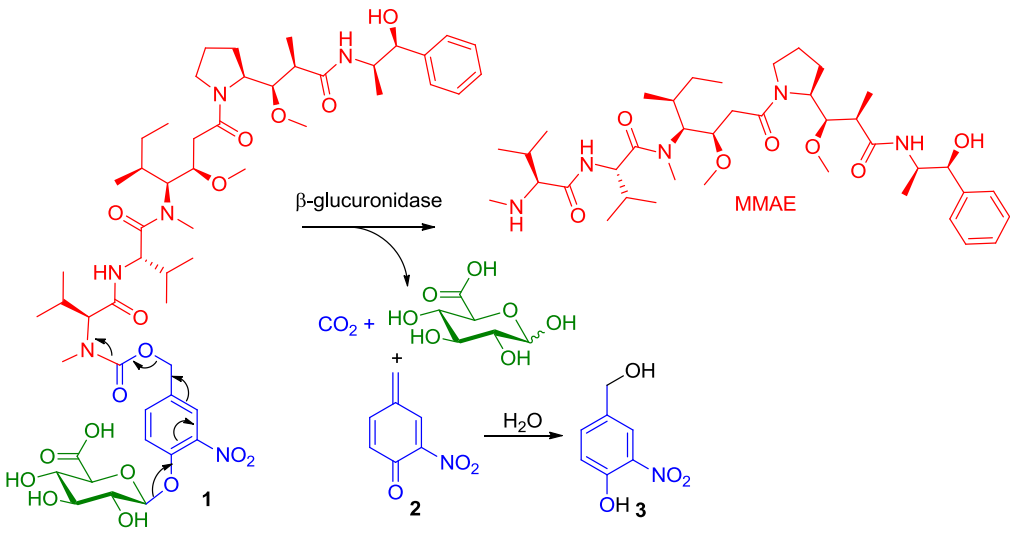


Fig. 1. Structure of prodrug **1** and its self-immolative mechanism upon β-glucuronidase activation.

amount of drug liberated at the tumor site is usually not sufficient to induce total and lasting remission of the tumor.

In order to overcome this drawback, we recently studied either dimeric [7] or heterodimeric [8] glucuronide prodrugs designed to release several drug units after a single enzymatic activation step. These self-immolative molecular systems were more toxic than the corresponding monomeric counterparts when activated with the same quantity of β-glucuronidase showing that this alternative may enhance the therapeutic efficacy of glucuronide prodrugs. Another approach proposed earlier by Tietze and coworkers relies on the targeting of highly potent duocarmycin analogs which are too toxic to be used in the course of standard chemotherapy [9]. In the presence of β-glucuronidase, the corresponding prodrugs were active at low picomolar concentrations making these compounds good candidates for *in vivo* experiments.

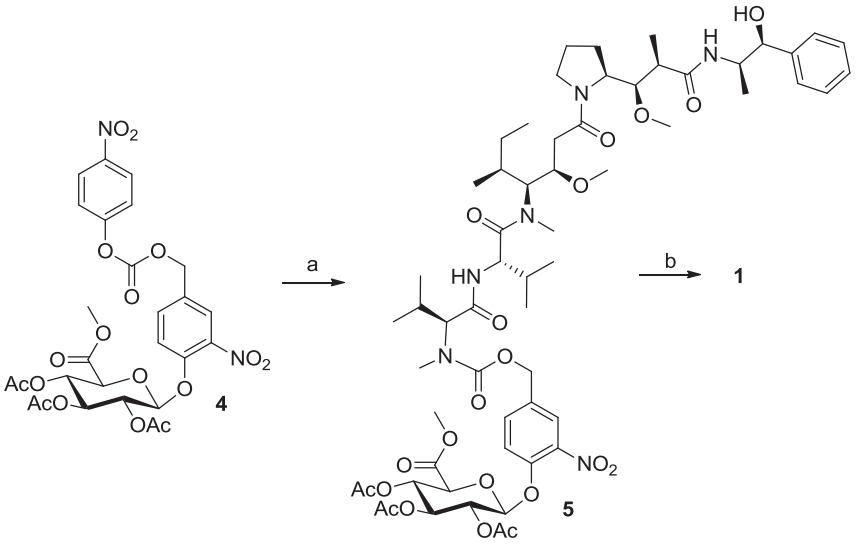
In this paper, we report the synthesis, *in vitro* and preliminary *in vivo* evaluations of the glucuronide prodrug **1** of monomethylauristatin E (MMAE [3], Fig. 1). MMAE is a potent inhibitor of tubulin polymerization with subnanomolar cytotoxic activity against a wide range of cancer cell lines. Moreover, MMAE has already shown a remarkable efficiency in human when targeted to cancer cells in the form of Brentuximab Vedotin. Thus, it seems that MMAE possesses all the main requirements to be targeted at the tumor site by the mean of a glucuronide prodrug.

Prodrug **1** includes a self-immolative linker [10] between the carbohydrate and the drug to allow an easy recognition of the glucuronide moiety by β-glucuronidase. With this design, enzymatic hydrolysis of the glycosidic bond will trigger the release of MMAE in a stringently controlled fashion *via* the mechanism depicted in Fig. 1.

2. Results and discussion

2.1. Chemistry

Prodrug **1** was prepared from the readily accessible activated carbonate **4** (Scheme 1) which represents an ideal platform for the rapid synthesis of glucuronide prodrugs. Indeed, this intermediate has been employed with success to introduce a wide variety of primary and secondary amine-containing drugs including



Scheme 1. Reagents and conditions: (a) MMAE, HOBt, DIPEA, DMF/pyridine 8/2, rt, 16 h, 68%; (b) LiOH (8.8 equiv.), MeOH, rt, 15 min, 78%.

doxorubicin [11], histone deacetylases inhibitors like CI-994 [12] or MS-275 [8], nornitrogen mustard [13] and cyclopamine [14]. Thus, MMAE was first coupled with **4** in the presence of 1-hydroxybenzotriazole and *N*,*N*-diisopropylethylamine in DMF to give the carbamate **5** in 68% yield. The full deprotection of the glucuronide moiety was then carried out using an excess of LiOH in methanol to afford the prodrug **1** which was further purified by preparative chromatography for biological evaluations (yield 78%, purity > 95%).

2.2. Stability and enzymatic hydrolysis

The stability of **1** was examined in bovine serum at 37 °C. After 48 h of incubation no decomposition was observed by HPLC indicating that prodrug **1** is highly stable under physiological conditions. Enzymatic hydrolysis of glucuronide **1** was then undertaken in phosphate buffer (0.02 M, pH 7, 37 °C) in the presence β-glucuronidase (Fig. 2). When incubated with the enzyme, prodrug **1** was rapidly cleaved leading to the clean release of MMAE together with the formation of the 4-hydroxy-3-nitrobenzyl alcohol **3** which resulted from the nucleophilic addition of water to the methylene quinone **2** (Fig. 1). This result confirmed that the disassembly of **1** proceeded through the self-immolative mechanism illustrated in Fig. 1. Furthermore, since the full expulsion of MMAE was accomplished in less than 40 min, the kinetics of drug release is compatible with a tumor-activated prodrug therapy. Indeed, in this approach the liberation of the active compound has to occur quickly after the enzymatic activation step in order to avoid the diffusion of the linker-drug intermediate outside of the tumor area. Thus, the fast elimination of MMAE from prodrug **1** should prevent the unselective release of the drug in healthy tissues.

Table 1
IC_{50} values (nM) for cell-growth inhibition assays.

	IC_{50} (nM)		
	MMAE	1	1 + βglu
A549	0.59 ± 0.14	23.7 ± 2.7	0.52 ± 0.15
KB	0.19 ± 0.03	20.8 ± 3.4	0.16 ± 0.04
MDA-MB-231	0.25 ± 0.03	20.9 ± 4.5	0.22 ± 0.09

2.3. Biological evaluations

2.3.1. Antiproliferative activity on tumor cell lines and primary cultures

Prodrug 1 was first evaluated for its antiproliferative activity against A549 (human lung adenocarcinoma), KB (human oral squamous carcinoma) and MDA-MB-231 (human breast adenocarcinoma) cells after four days treatment. When incubated with β-glucuronidase, prodrug 1 induced a dramatic cytotoxic effect with IC_{50} ranging from 0.16 to 0.52 nM. These values were similar to that recorded with MMAE, indicating the efficient release of the free drug in the culture medium (Table 1). On the other hand, in the absence of the activating enzyme, prodrug 1 was 40–110 fold less toxic than the drug. These results showed that the derivatization of MMAE in the form of prodrug 1 markedly reduced its cytotoxicity. As expected, the hydrophilicity imparted by the glucuronide trigger limited passive cellular uptake and further intracellular activation of the prodrug by lysosomal β-glucuronidase.

The antiproliferative activity of prodrug **1** was also tested on primary cultures of five patients who underwent surgical resection for primary non-small cell lung cancer. Cells were treated with 0.5

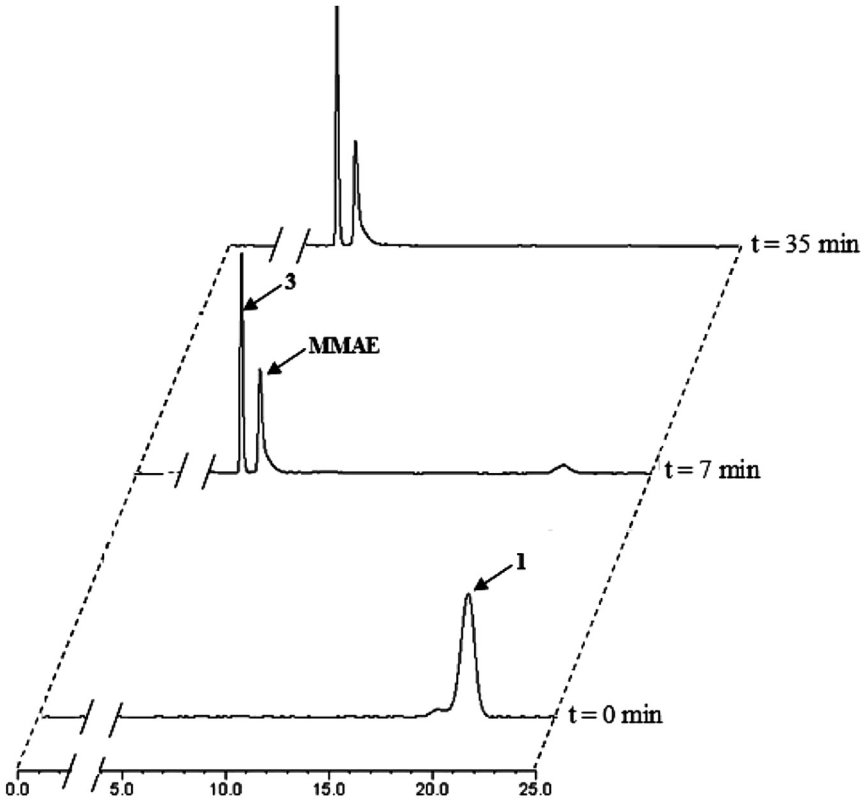


Fig. 2. Enzymatic hydrolysis of prodrug **1** with *E. coli* β-glucuronidase in phosphate buffer (0.02 M, pH 7) at 37 °C.

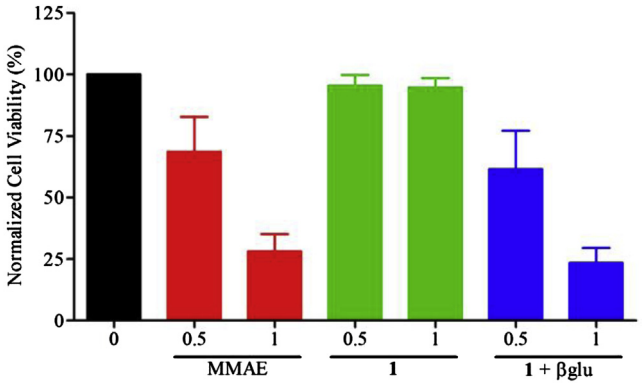


Fig. 3. Cytotoxicity of prodrug 1 on primary cultures of patients with lung cancer (mean of 6 patient samples).

or 1 nM of MMAE or **1**, with or without β-glucuronidase. The mean of cell viability for the five patient samples is reported in Fig. 3. Thus, while the prodrug did not affect the viability of cells when incubated alone at the two tested doses, addition of the enzyme in the medium restored the cytotoxicity of MMAE. It is worth mentioning here that upon β-glucuronidase activation, prodrug **1** produced a strong cytotoxic effect at a dose as low as 1 nM, killing about 75% of the cells.

2.3.2. In vivo experiments

The *in vivo* evaluation of prodrug **1** was conducted in C57BL/6 mice bearing a subcutaneous murine Lewis Lung Carcinoma (LLC). The antitumor efficacy of **1** was compared to that of MMAE and HMR 1826, a well-known glucuronide prodrug of doxorubicin [6b]. The animals received three i.v. injections of MMAE (0.1 mg kg^{-1}, 0.14 μM kg^{-1}), prodrug 1 (0.5 mg kg^{-1}, 0.46 μM kg^{-1}) or HMR 1826 (100 mg kg^{-1}, 109 μM kg^{-1}) at days 7, 11 and 14 after transplantation. On day 20, mice were euthanized and tumor weights were measured. As shown in Fig. 4, MMAE, 1 and HMR 1826 induced a similar antitumor effect with tumor weights of 0.785 g, 0.798 g and 0.753 g respectively. In comparison, the tumor weight recorded in untreated animals was 1.5-fold higher (1.176 g) therefore demonstrating the therapeutic benefit brought by the tested compounds. While the treatments with glucuronide **1** or HMR 1826 limited the tumor growth in a comparable fashion, the prodrug of MMAE was administered at a 237-fold lower dose in these experiments. Moreover, prodrug **1** was well tolerated without any sign of overt toxicity at the tested dose (see the Supporting Information). Taken all together, these results confirmed that glucuronide prodrugs of highly potent cytotoxics such as **1** could be a valuable alternative to enhance the efficiency of this targeting strategy. Indeed, as a significant antitumor effect can be obtained at low concentrations, the efficiency of prodrug **1** may not be limited by the turnover of β-glucuronidase in the tumor microenvironment. Further *in vivo* assays with higher doses of prodrug **1** will be carried out shortly in order to verify this hypothesis.

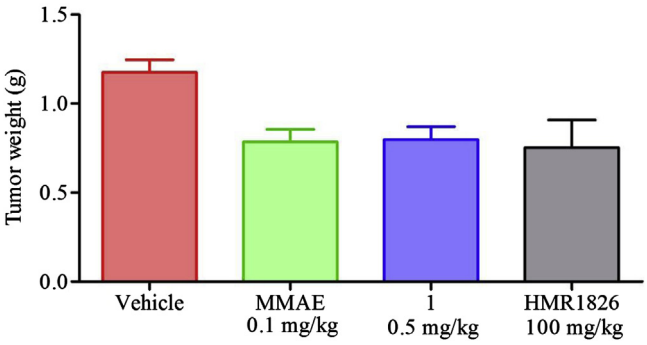


Fig. 4. Tumor weights after treatment with the indicated compounds (6 mice per group).

3. Conclusion

In summary, we synthesized a glucuronide prodrug of the potent antimitotic agent MMAE which can be selectively activated by β-glucuronidase overexpressed in the microenvironment of numerous tumors. This compound was stable under physiological conditions and exhibited a reduced toxicity compared to the free drug. In the presence of the activating enzyme, the prodrug led to the clean release of MMAE thereby restoring its cytotoxicity. *In vivo* experiments showed that this β-glucuronidase-responsive targeting system was well tolerated in mice at a dose inducing a significant antitumor effect. Furthermore, the prodrug of MMAE was as efficient as its doxorubicin analog when administered at a 237-fold lower dose. Thus, all these data suggest that this new glucuronide possesses the necessary prerequisites for further *in vivo* investigation in the course of a tumor targeting strategy.

4. Experimental

4.1. General chemistry methods

All reactions were performed under N_2 atmosphere. Unless otherwise stated, solvents used were of HPLC quality. Chemicals were of analytical grade from commercial sources and were used without further purification. ^1H and ^{13}C NMR were performed on an Avance 400 Bruker spectrometer. The chemical shifts are expressed in part per million (ppm) relative to TMS (δ = 0 ppm) and the coupling constant J in hertz (Hz). NMR multiplicities are reported using the following abbreviations: b = broad, s = singlet, d = doublet, t = triplet, q = quadruplet, m = multiplet. High resolution ESI mass spectrometry was carried out by the CRMPO (Centre Régional de Mesure Physiques de l'Ouest), at the University of Rennes 1. The reaction progress was monitored on precoated silica gel TLC plates Macherey-Nagel ALUGRAM® SIL G/UV_{254} (0.2 mm silica gel 60 Å). Spots were visualized under 254 nm UV light and/or by dipping the TLC plate into a solution of 3 g of phosphomolibdic acid in 100 mL of ethanol followed by heating with a heat gun. Flash column chromatography was performed using Macherey-Nagel silica gel 60 (15–40 μm). Analytical RP-HPLC was carried out on a Dionex Ultimate 3000 system equipped with a UV/visible variable wavelength detector and with a reverse-phase column chromatography Acclaim® (C18, 250 × 4.6 mm, 5 mm, 120 Å) at 30 °C and 1 mL min^{-1}. Gradient eluent was composed of A (0.2% TFA in water) and B (CH_3CN). Method **A**: linear gradient beginning with A/B 80/20 v/v, reaching 0/100 v/v within 30 min. Method **B**: isocratic gradient with A/B 60/40 v/v, 30 min. Preparative reverse phase HPLC for **1** was performed with a VWR LaPrep system. Solvent flow 4 mL min^{-1} was applied to a semi-preparative column ACE 5® (C18, 100 × 10 mm). Gradient eluent was composed of A (0.02% TFA in water) and B (CH_3CN). Method: linear gradient beginning with A/B 80/20 v/v, reaching A/B 0/100 v/v within 20 min. All chromatograms were recorded at 254 nm.

4.2. Synthesis and characterization of described compounds

*4.2.1. Synthesis and characterization of compound **5***

MMAE (25 mg, 0.035 mmol) and carbonate **4** (23 mg, 0.035 mmol) were dissolved in dry DMF (800 μL) and pyridine

(200 μL). HOBt (4.7 mg, 0.035 mmol) and DIPEA (7.3 μL, 0.042 mmol) were added. The mixture was stirred at room temperature. After 48 h, solvents were removed under reduced pressure and the crude material was purified by preparative TLC over silica gel (CH_2Cl_2/MeOH, 97/3) to afford **5** (29 mg, 68%) as a white solid (retention time = 22.6 min for HPLC analysis (method A)).

^{1}H NMR (400 MHz, $CDCl_3$) 0.73–1.02 (m, 23H), 1.20–1.39 (m, 4H), 1.85 (m, 3H), 1.95–2.11 (m, 12H), 2.20–2.50 (m, 4H), 2.85–3.10 (m, 6H), 3.30–3.41 (m, 7H), 3.51 (m, 1H), 3.74 (s, 3H), 3.85 (m, 1H), 3.96–4.25 (m, 6H), 4.67–4.76 (m, 2H), 4.96 (m, 1H), 5.09–5.38 (m, 6H), 6.48–6.55 (m, 1H), 7.25 (m, 1H, masked by $CDCl_3$ residual signal), 7.35 (m, 5H), 7.53 (m, 1H), 7.79 (m, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) 11.0, 14.0, 14.2, 14.5, 15.5, 16.1, 17.4, 17.6, 18.7, 19.4, 20.6, 20.7, 22.0, 23.5, 25.0, 25.1, 25.9, 26.3, 29.5, 29.8, 31.1, 32.0, 33.5, 37.8, 45.0, 47.9, 51.7, 53.2, 53.5, 54.0, 58.1, 60.2, 61.0, 65.4, 65.7, 68.8, 70.3, 71.2, 72.7, 75.9, 78.6, 82.1, 99.9, 120.2, 124.5, 125.2, 126.4, 127.4, 128.2, 128.4, 133.2, 133.4, 133.9, 141.3, 148.8, 149.0, 156.9, 166.8, 169.3, 169.4, 169.8, 170.1, 174.7. HRMS (ESI) $[M + Na]^+$ m/z 1251.5903 (calcd. for $C_{60}H_{88}N_6O_{21}Na$: 1251.58947).

4.2.2. Synthesis and characterization of prodrug **1**

5 (29 mg, 0.024 mmol) was dissolved in MeOH (1.6 mL). The mixture was cooled at 0 °C and a solution of lithium hydroxide monohydrate (8.7 mg, 0.208 mmol) in water (1.6 mL) was added dropwise. The mixture was stirred for 15 min, hydrolyzed with IRC-50 acidic resin, filtrated and concentrated in vacuo. High degree of purity for **1** was obtained using preparative-reverse phase HPLC (20 mg, 78%, purity > 95%). Retention time 15.9 min (for HPLC conditions see HPLC analysis – Method A).

^{1}H NMR (400 MHz, DMSO d6) 0.7–1.07 (m, 25H), 1.23 (m, 1H), 1.50 (m, 1H), 1.77 (m, 2H), 1.94–2.14 (m, 2H), 2.2–2.3 (m, 1H), 2.4–2.7 (m, 5H masked by DMSO d6 residual signal), 2.8–2.9 (m, 3H), 2.97–3.3 (m, 13H), 3.56 (m, 9H masked by H_2O residual signal), 3.97 (m, 3H), 4.26 (m, 0.6H), 4.43 (m, 1H), 4.68 (m, 0.5H), 5.15 (m, 3H), 7.17 (m, 1H), 7.27 (m, 4H), 7.4 (m, 1H), 7.64 (m, 1H), 7.88 (m, 1H), 8.15 (m, 0.5H), 8.4 (m, 0.5H). ^{13}C NMR (100 MHz, DMSO d6) 10.81, 14.21, 14.25, 15.38, 15.52, 15.65, 15.79, 15.86, 18.82, 18.91, 19.07, 19.17, 19.3, 23.7, 23.8, 24.64, 24.84, 25.63, 25.73, 25.88, 30.26, 30.39, 31.92, 32.09, 46.6, 49.51, 49.65, 50.24, 54.62, 54.74, 56.54, 57.5, 57.69, 58.54, 58.68, 60.65, 60.68, 61.45, 63.6, 63.7, 65.17, 65.24, 71.53, 73.3, 75.35, 75.82, 75.96, 76.37, 100.48, 117.47, 124.27, 124.3, 124.46, 127.02, 128.25, 128.37, 131.7, 131.8, 140.4, 144.17, 147.05, 147.11, 149.16, 150.36, 151.87, 169.24, 170.44, 174.02; HRMS (ESI) $[M - H]^-$ m/z 1087.5444 (calcd. for $C_{53}H_{79}N_6O_{18}$: 1087.54564).

4.3. Stability

Compound **1** (0.5 mg) was incubated at 37 °C in bovine serum (0.2 mL). Aliquots at t = 0 and t = 48 h (50 μL) were withdrawn from the medium, poured into cold MeOH (100 μL) to precipitate the proteins and cooled on ice. After 30 min, the sample was centrifuged (9000 rpm, 5 min) and the supernatant analyzed by analytical HPLC using Method B. HPLC analysis showed no detectable degradation of compound **1** during 48 h under these conditions.

4.4. Enzymatic hydrolysis

Enzymatic hydrolysis was carried out with commercially available β-glucuronidase from *Escherichia coli* (purchased from Sigma–Aldrich reference: G8162). Prodrug **1** (0.1 mg mL^{-1}) was incubated with β-glucuronidase (133 U mL^{-1}) in phosphate buffer (0.02 M, pH 7) at 37 °C. Aliquots of 20 μL were taken at indicated time and analyzed by HPLC using Method B. Retention time for compounds **1**, **3** and MMAE are 20.7, 5.1 and 6.0 min respectively.

4.5. Cell culture

KB (human oral squamous carcinoma), MDA-MB-231 (human breast adenocarcinoma) and A549 (human lung carcinoma) cells were grown in RPMI 1640 (Invitrogen) supplemented by 10% fetal bovine serum and 1% Penicillin/Streptomycin (Lonza) in a humidified incubator at 37 °C and 5% CO_2.

4.6. Cell viability

The Cell Proliferation Kit II (XTT; Roche) was used to assess cell viability. This assay is based on the cleavage of XTT by metabolic active cells resulting in the production of an orange formazan dye quantified by spectrophotometry. Assays were carried out essentially as described by the manufacturer. Briefly, 2×10^3 tumor cells/well in a 96-well plate were plated. Cells were cultured for 24 h before adding the compound at the indicated concentration in the culture media. After 4 days of treatment, 50 μL of the XTT labeling mixture were added per well. Cells were further incubated for additional 4 h at 37 °C before determination of the absorbance at 490 nm. Experiments were performed 4–6 times.

4.7. Primary cultures of human bronchial epithelial cells

Fragment of human lung carcinoma were obtained from five Caucasian patients who underwent curative operation for primary non-small cell lung cancer. Curative operation was defined as the complete removal of the ipsilateral hilar and mediastinal lymph nodes together with the primary tumor including negative bronchial margins. Patients did not receive radiotherapy or chemotherapy before surgery. There were three males and two females, age: 60 ± 13 years. All tumor patients are adenocarcinoma, stade I or IIa. Tissues were received and processed within 2 h after surgery. All procedures were performed in compliance with the French legislation. All resected lobes and lymph nodes were investigated histopathologically. The tumor cells were microscopically dissected from their 5-μm sections for DNA extraction. Direct sequencing from EGFR exon 18, exon 19, exon 20, and exon 21 was performed. Bronchial epithelial cells were harvested using enzymatic isolation procedures to establish primary cultures [15]. Fragments of bronchi were incubated 24 h at 4 °C with 0.1% protease and 0.01% deoxyribonuclease in DMEM/Ham's F-12 medium. Enzymatic digestion was neutralized by 10% fetal calf serum; cells were then centrifuged at 700 rpm for 7 min at room temperature. Cells were cultured in DMEM/Ham's F-12 medium supplemented with: 5 μg mL^{-1} insulin, 7.5 μg mL^{-1} transferrin, 10^{-6} M hydrocortisone, 2 μg mL^{-1} endothelial cell growth supplement, 25 ng mL^{-1} epithelial growth factor, 3×10^{-8} M triiodothyronine, 2.5 mM L-glutamine, and 100 IU mL^{-1} penicillin and 100 μg mL^{-1} streptomycin. Cells were then treated with 0.5 or 1 nM MMAE or prodrug **1** for 4 days. Cell viability was quantified using XTT kit as previously described.

4.8. In vivo studies

C57BL/6 mice purchased from Janvier were the only animals used in this study. All experimental procedures involving animals were carried out in accordance with the guidelines of the French Agriculture and Forestry Ministry (decree 87849) and of the European Communities Council Directive (2010/63/UE). 5×10^5 LLC tumor cells mixed with Matrigel (BD Biosciences) were transplanted subcutaneously into the left flank region of the 10-weeks old mice (day 0). Mice were randomly distributed in 3 experimental groups of 6 mice. The mice were then treated intravenously on days 7, 11 and 14 with either 5% DMSO in 1X PBS buffer (vehicle), MMAE (0.1 mg kg^{-1}, 0.14 μM kg^{-1}), prodrug 1 (0.5 mg kg^{-1}, 0.46 μM kg^{-1}) or HMR 1826

(100 mg kg^{-1}, 109 μM kg^{-1}) diluted as in 5% DMSO/PBS. On study day 20, mice were euthanized and tumor weights were measured. Mice were weighted every 2 days during the time of the experiment.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.ejmech.2013.06.037.

References

[1] F. Kratz, I.A. Müller, C. Ryppa, A. Warnecke, Chem. Med. Chem. 3 (2008) 20–53.
[2] S.C. Alley, N.M. Okeley, P.D. Senter, Curr. Opin. Chem. Biol. 14 (2010) 529–537.
[3] S.O. Doronina, B.E. Toki, M.Y. Torgov, B.A. Mendelsohn, C.G. Cerveny, D.F. Chace, R.L. DeBlanc, R.P. Gearing, T.D. Bovee, C.B. Siegall, J.A. Francisco, A.F. Wahl, D.L. Meyer, P.D. Senter, Nat. Biotechnol. 21 (2003) 778–784.
[4] (a) T.A. Connors, M.E. Whisson, Nature 210 (1966) 866–867;
(b) K. Bosslet, J. Czech, D. Hoffmann, Tumor Targeting 1 (1995) 45–50.
[5] Tietze was the first who proposed to target this enzyme by the mean of glucuronide prodrugs: L.F. Tietze, R. Seele, B. Leiting, T. Krach Carbohydr. Res. 180 (1988) 253–262.
[6] (a) P.H.J. Houba, E. Boven, C.A.M. Erkelens, R.G.G. Leenders, J.W. Scheeren, H.M. Pinedo, H.J. Haisma, Br. J. Cancer 78 (1998) 1600–1606;
(b) K. Bosslet, R. Straub, M. Blumrich, J. Czech, M. Gerken, B. Sperker, H.K. Kroemer, J.-P. Gesson, M. Koch, C. Monneret, Cancer Res. 58 (1998) 1195–1201;
(c) R. Woessner, Z. An, X. Li, R.M. Hoffman, R. Dix, A. Bitonti, Anticancer Res. 20 (2000) 2289–2296;
(d) P.H.J. Houba, E. Boven, I.H. Van Der Meulen-Muileman, R.G.G. Leenders, J.W. Scheeren, H.M. Pinedo, H.J. Haisma, Br. J. Cancer 84 (2001) 550–557;
(e) T.Y. Juan, S.R. Roffler, H.S. Hou, S.M. Huang, K.C. Chen, Y.L. Leu, Z.M. Prijovich, C.P. Yu, C.C. Wu, G.H. Sun, T.L. Cha, Clin. Cancer Res. 15 (2009) 4600–4611;
(f) S. Chevalier, C. Goupille, K. Mahéo, I. Domingo, C. Dussiau, B. Renoux, P. Bougnoux, S. Papot, Clin. Lipidol. 5 (2010) 233–243;
(g) T. Legigan, J. Clarhaut, B. Renoux, I. Tranoy-Opalinski, A. Monvoisin, J.-M. Berjeaud, F. Guilhot, S. Papot, J. Med. Chem. 55 (2012) 4516–4520.
[7] M. Grinda, J. Clarhaut, B. Renoux, I. Tranoy-Opalinski, S. Papot, Med. Chem. Commun. 3 (2012) 68–70.
[8] M. Grinda, J. Clarhaut, I. Tranoy-Opalinski, B. Renoux, A. Monvoisin, L. Cronier, S. Papot, Chem. Med. Chem. 6 (2011) 2137–2141.
[9] (a) L.F. Tietze, K. Schmuck, H.J. Schuster, M. Müller, I. Schuberth, Chem. Eur. J. 17 (2011) 1922–1929;
(b) L.F. Tietze, H.J. Schuster, K. Schmuck, I. Schuberth, F. Alves, Bioorg. Med. Chem. 16 (2008) 6312–6318.
[10] (a) S. Papot, I. Tranoy, F. Tillequin, J.-C. Florent, J.-P. Gesson, Curr. Med. Chem. Anti-Cancer Agents 2 (2002) 155–185;
(b) I. Tranoy-Opalinski, A. Fernandes, M. Thomas, J.-P. Gesson, S. Papot, Anti-Cancer Agents Med. Chem. 8 (2008) 618–637.
[11] J.C. Florent, X. Dong, G. Gaudel, S. Mitaku, C. Monneret, J.P. Gesson, J.C. Jacquesy, M. Mondon, B. Renoux, S. Andrianomenjanahary, S. Michel, M. Koch, F. Tillequin, M. Gerken, J. Czech, R. Straub, K. Bosslet, J. Med. Chem. 41 (1998) 3572–3581.
[12] M. Thomas, J. Clarhaut, I. Tranoy-Opalinski, J.-P. Gesson, J. Roche, S. Papot, Bioorg. Med. Chem. 16 (2008) 8109–8116.
[13] S. Papot, D. Combaud, K. Bosslet, M. Gerken, J. Czech, J.-P. Gesson, Bioorg. Med. Chem. Lett. 10 (2000) 1835–1837.
[14] F. Hamon, B. Renoux, C. Chadéneau, J.-M. Muller, S. Papot, Eur. J. Med. Chem. 45 (2010) 1678–1682.
[15] (a) S. Blouquit, H. Morel, J. Hinnrasky, E. Naline, E. Puchelle, T. Chinet, Am. J. Respir. Cell Mol. Biol. 27 (2002) 503–510;
(b) L. Dannhoffer, S. Blouquit-Laye, A. Renier, T. Chinet, Am. J. Respir. Cell Mol. Biol. 40 (2009) 717–723.