A Novel Tumor-Activated Prodrug Strategy Targeting Ferrous Iron Is Effective in Multiple Preclinical Cancer Models


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Current Literature Seminar
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Iron and Cancer

Iron enables the function of
- Vital iron and haem-containing enzymes involved in respiratory complexes (mitochondrial enzymes)
- Enzymes involved in DNA synthesis and cell cycle
- Detoxifying enzymes such as peroxidase and catalase

- Iron is essential for cell replication, metabolism and growth
Iron (Uptake and Efflux) in Normal vs. Cancer Cells

Fe^{2+} + H_2O_2 \rightarrow Fe^{3+} + HO^- + HO^-

Fe^{3+} + H_2O_2 \rightarrow Fe^{2+} + HOO^- + H^+
Fe(II)-Dependent Drug Delivery

- Labile Fe(II) promotes Fenton chemistry
- Fenton reaction of a peroxidic prodrug coupled to release drug payloads

- Drug species can be conjugated via an amine or alcohol function, potentially allowing the intrinsic bioactivity and/or toxicity of the drug species to be blocked before Fe(II) dependent release at the desired side of action.
Synthesis of Microtubule Inhibitor

\[ \text{LDA, TMSCHN}_2, \text{THF}, -78 ^\circ \text{C}, 56\% \]

\[ \text{EtMgCl, THF} \]
\[ \text{N}_3, 34\% \]

\[ \text{H}_2, \text{Pd/C}, \text{MeOH}, 98\% \]

\[ \text{COC}_2, 70 ^\circ \text{C} \]
\[ \text{toluene, sealed tube, 6 - 16 h} \]

\[ \text{O}_2, \text{CCl}_4, 25 ^\circ \text{C}, 2 \text{ h} \]

\[ \text{TBAF, THF, 0 ^\circ \text{C}, rt, 24 \text{ h}} \]

\[ \text{KOH, Me}_2\text{SiO, rt} \]

\[ \text{81-91\%} \]

\[ \text{df = 90:10} \]

\[ \text{13, R = H} \]

\[ \text{7, R = Me} \]

\[ \text{9, (X=O) 59\% over 2 steps} \]

\[ \text{10, (X=O) 20\% over 2 steps} \]
Design, Synthesis and Validation in Cell Culture of a Microtubule Toxin

- Drug release is both efficient and peroxide dependent
Cytotoxicity in a Panel of Cancer Cell Lines

“$E_{50}$ ratio” → Normalizing the activity of conjugate 2 to that of its cytotoxic payload 1 to compare efficiency of payload release from 2 across different cell lines
In Vivo PK/PD Studies of Duocarmycin Conjugate

**Duocarmycin** isolated from Streptomyces bacteria. Known for extreme cytotoxicity. Extremely potent antitumor antibiotics

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<th></th>
<th>T1/2 [h]</th>
<th>Clearance [mL min⁻¹ kg⁻¹]</th>
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<td>20.4</td>
<td>31.3</td>
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b. **PC-3**

![Graph showing the effect of different compounds on PC-3 cell viability](image)

- **MDA-MB-231**

![Graph showing the effect of different compounds on MDA-MB-231 cell viability](image)
PK Profile and *In Vivo* tolerability

Plasma concentrations

![Graph showing plasma concentrations over time](image)

Mouse liver samples

![Image of mouse liver samples](image)
Xenograft Studies

MDA-MB-231 xenograft bearing female SCID-beige mice

IP administration on Q4d schedule (3 total doses)
Conclusion

- Trioxolane-mediated Fe(II)-dependent drug delivery acts as a new approach for cell/tissue selective drug targeting

- Two prototypical trioxolane drug conjugates bearing cytotoxins with distinct mechanisms of cellular toxicity

- Confirmed that intrinsic cytotoxicity of these agents can be decreased in conjugated forms (and yet fully realized following cell or tumor selective release at their intended side of action)
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<td>P</td>
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<td>Phosphorus 30.974</td>
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<td>Dysprosium 162.50</td>
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Hydroxide, Hydroxide, Hydroxide!

Merry Christmas!