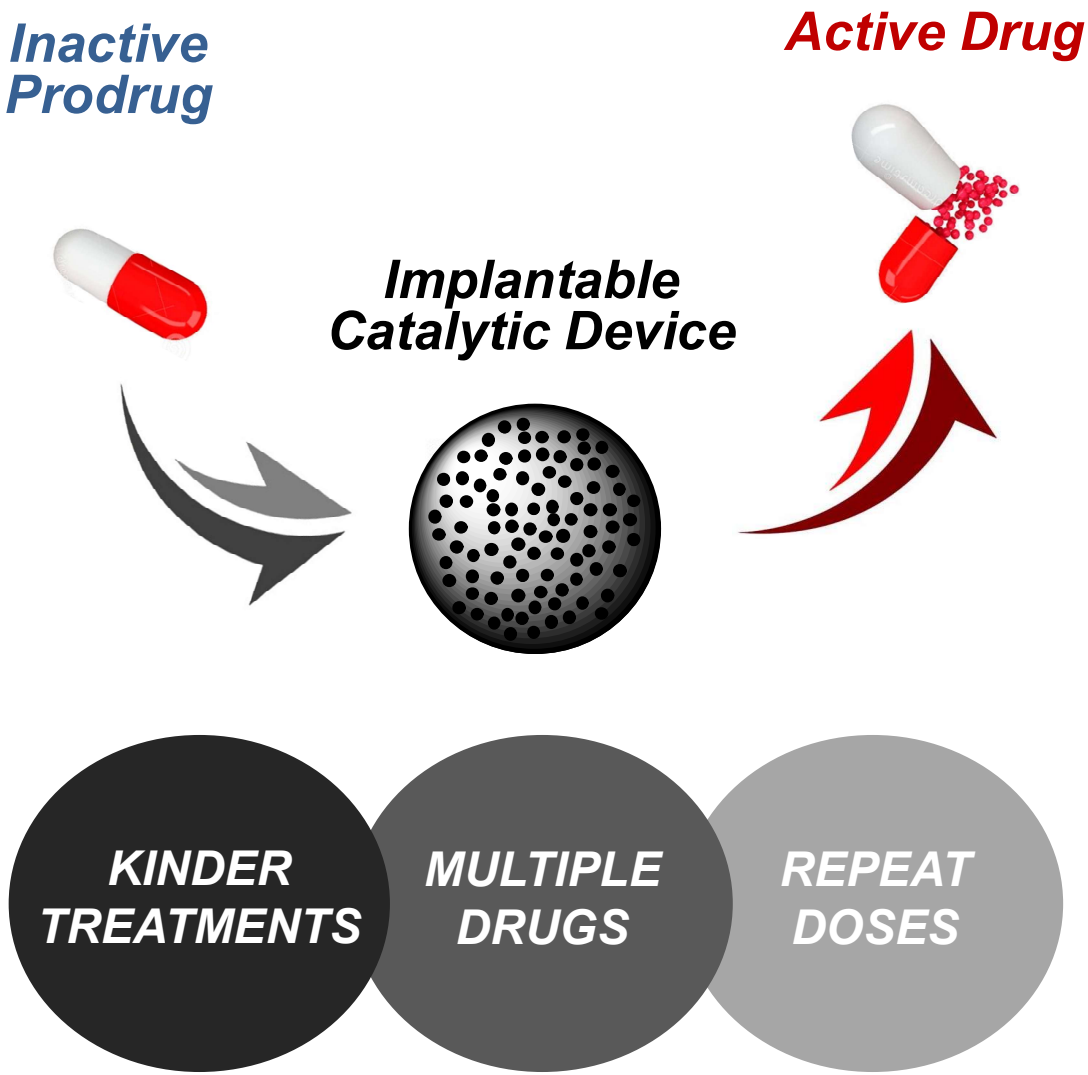
**AGENDA**

| | |
|---|---|
| 09:00 | Introduction |
| 09.15 | Lecture 1 (Historical overview of prodrugs) |
| 10.45 | Coffee Break |
| 11.00 | Lecture 2 (Bioorthogonal prodrugs 1) |
| 12:30 | Lunch |
| 13:30 | Lecture 3 (Bioorthogonal prodrugs 2) |
| 15:00 | Coffee Break |
| 15:30 | Lecture 4 (Challenges to progress metal-activated prodrugs into the clinic) |
| 17:00 | Go to hotel |
| 19:00 | Dinner (TBC) |

# Bioorthogonal Prodrugs 2

# Implants that make drugs inside your body



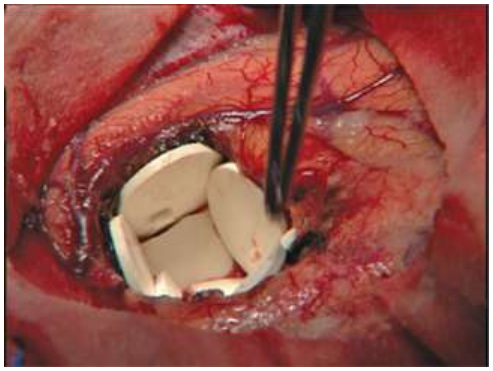
# Glioblastoma. An incurable disease

- ***Glioblastoma multiforme is the most frequent and deadly form of brain cancer***
- ***Locally infiltrates into the healthy tissue >> <u>surgery is not enough</u>***
- ***Chemo / radiotherapy applied to control cancer remnants***
- ***Survival from diagnosis with the best available treatments is merely 14 months***

GLIADEL® WAFER
(carmustine implant)

***Releases carmustine in the brain***

***Modest 2-month survival advantage***

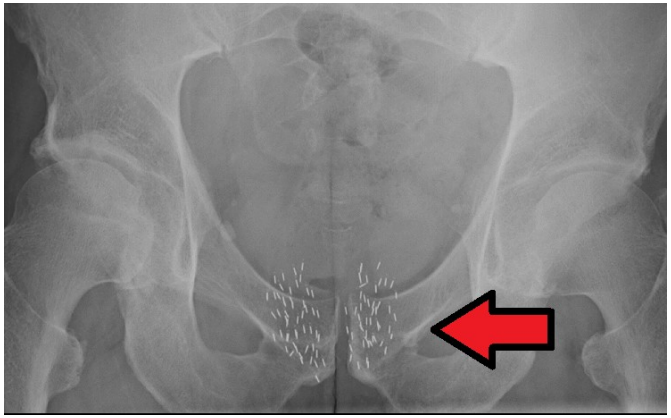


***POS: Targets local recurrence***
***CONS: increases risk of infection / limited cargo***

# Early Prostate Cancer. Untreated tumour

- ***50-70% of newly diagnosed cases of prostate cancer will fall within this category***
- ***Managed with <u>active surveillance</u> due to adverse effects of current therapies, leading to increased mortality risk***
- ***Available treatments: surgery and radioactive seeds into the prostate tumour***

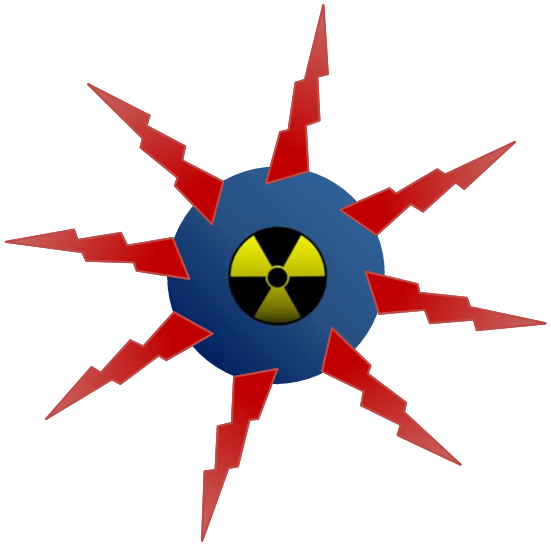
***Radiation is delivered directly into the prostate cancer. Healthy tissue nearby gets reduced dose***



***POS: targets local cancer growth***
***CONS: lacks prolonged activity***

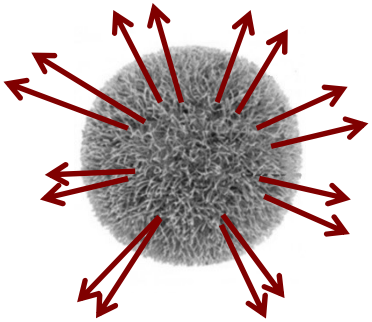
# Focal Therapies in the Clinic

***BRACHYTHERAPY***



- ***Radioactive seeds***
- ***Well established surgical insertion***
- ***Prostate cancer***
- ***One radionuclide // Limited half-life***

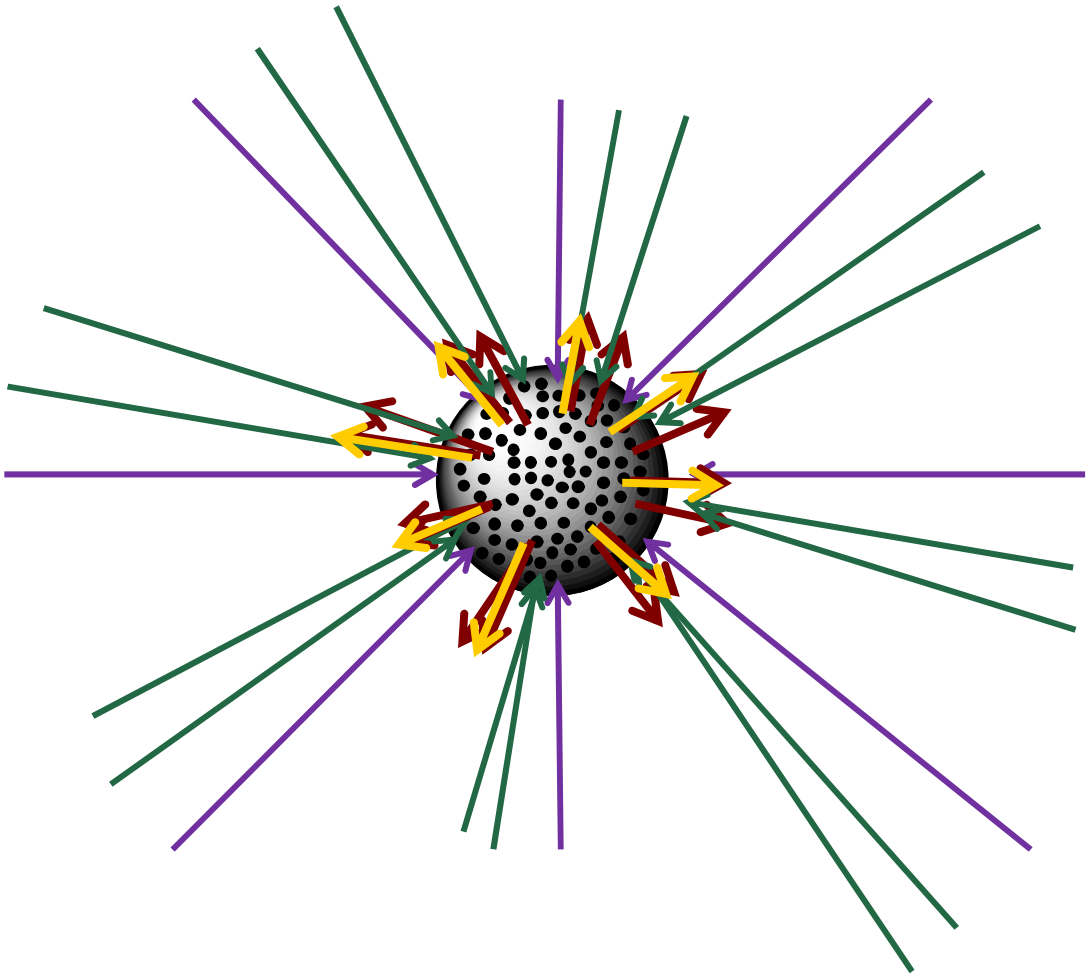
***CONTROLLED DRUG RELEASE***



- ***Carmustine wafers***
- ***Well established implantation***
- ***Glioblastoma multiforme***
- ***One drug // Limited cargo***

# FOCAL MULTIDOSE CHEMOTHERAPY

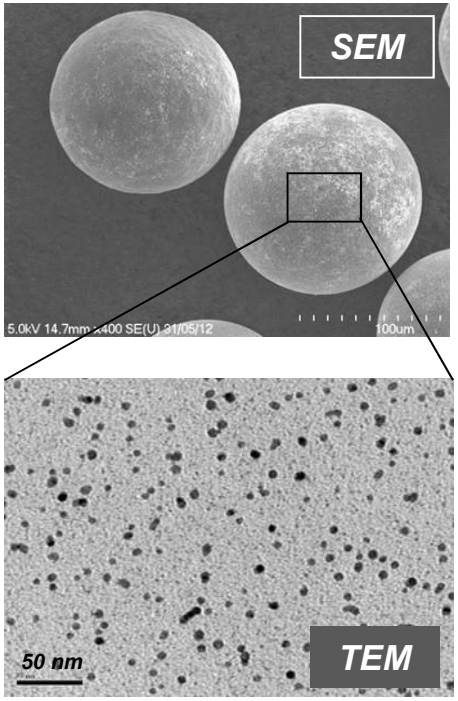
- *Catalyst*
- *Safe*
- *Palladium*
- *Gold*
- *Implantation*
- *etc.*

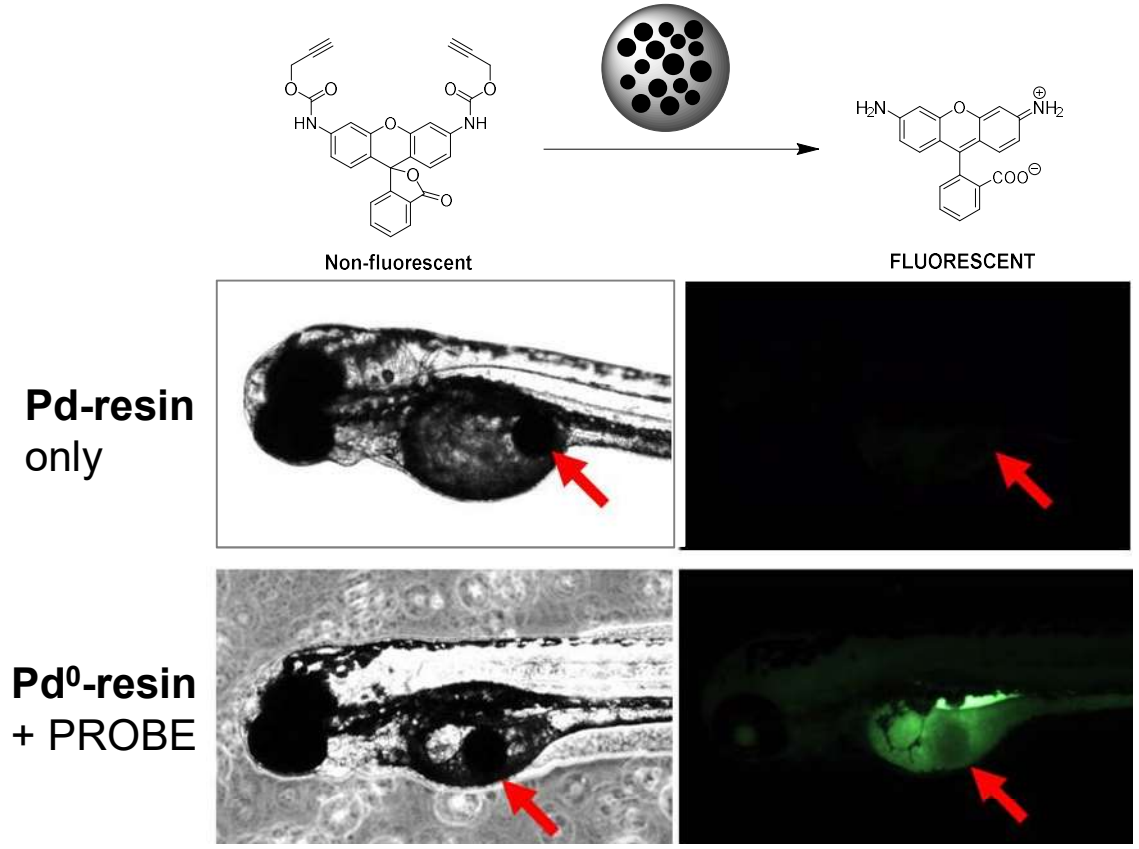


- *Doxorubicin*
- *5FU*
- *Floxuridine*
- *Vorinostat*
- *Irinotecan*
- *etc.*

# Heterogeneous Pd catalysis in vivo

***SOLID SUPPORT: Tentagel resins*** *(used in conventional solid-phase synthesis)*




*Fluorescent signal from the area surrounding the $Pd^0$-resin in the yolk sac confirm that the palladium-functionalized device is catalytically active in vivo*

*Nat. Commun.* **2014**, 5, 3277

# How to make a bioorthogonal prodrug

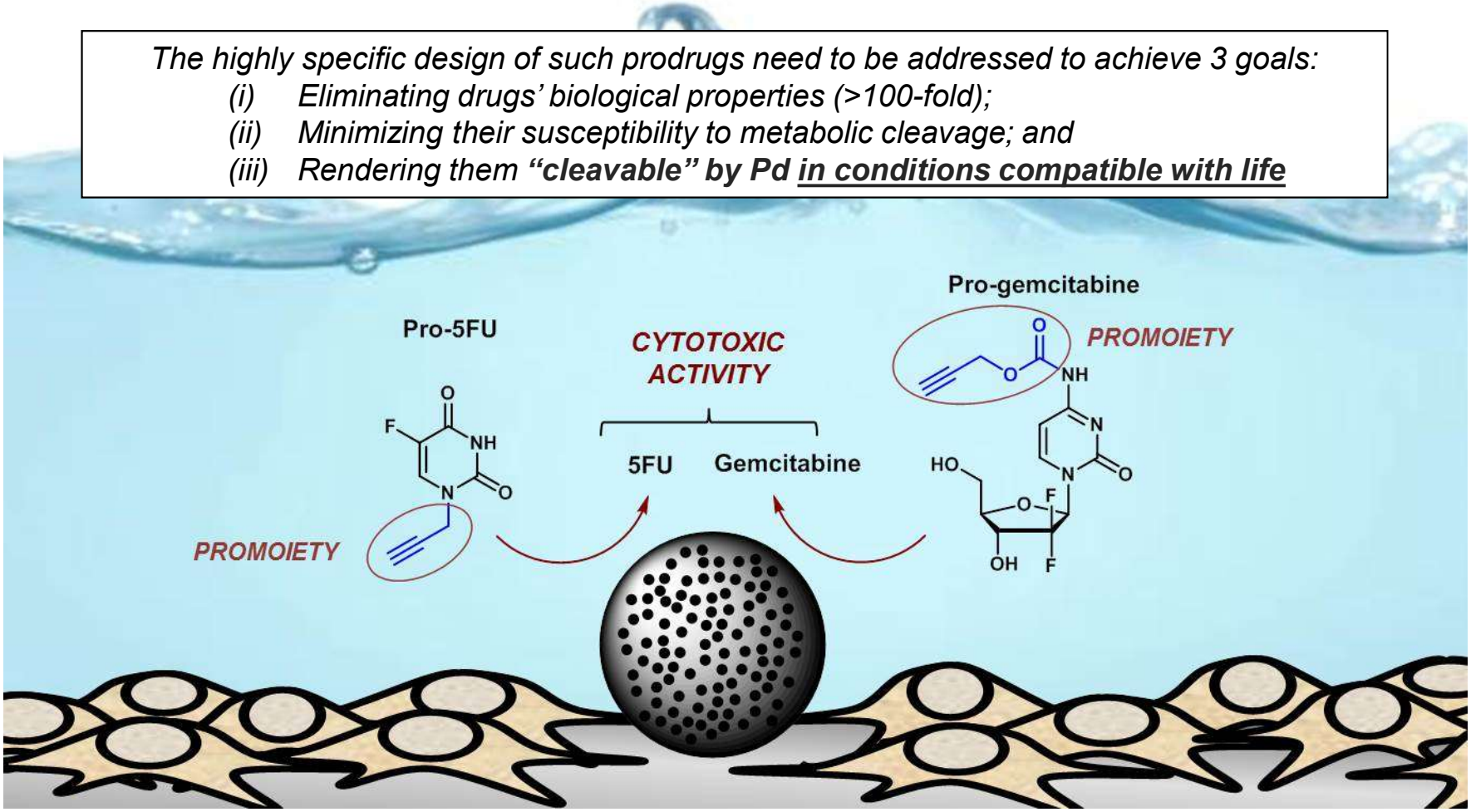
***OBJECTIVE:*** *Control the activation of prodrugs* ***exclusively*** *by implant-localized palladium catalysis*

*The highly specific design of such prodrugs need to be addressed to achieve 3 goals:*

*(i) Eliminating drugs' biological properties (>100-fold);*

*(ii) Minimizing their susceptibility to metabolic cleavage; and*

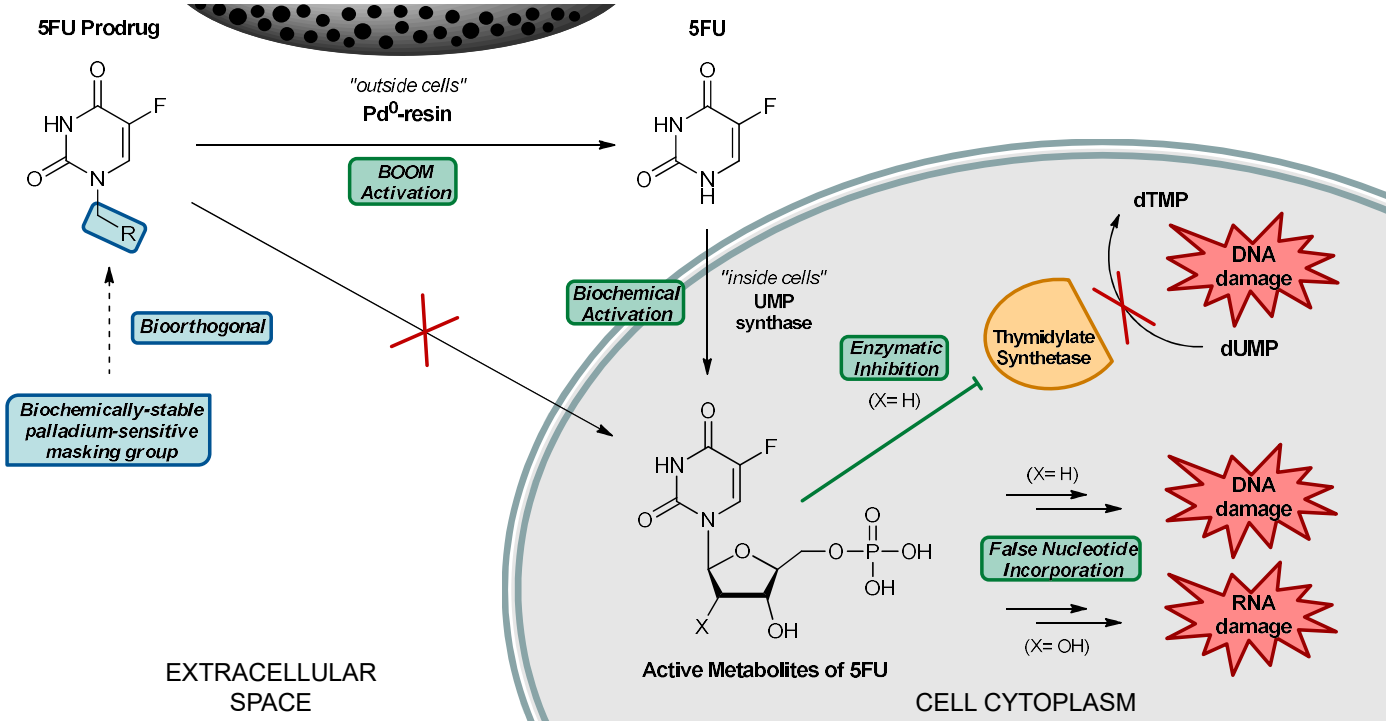
*(iii) Rendering them* ***"cleavable" by Pd*** ***<u>in conditions compatible with life</u>***




*Nat Commun* **2014**, 5, 3277 *J Med Chem* ***2014***, 57, 5395

# 5FU Prodrug Design

***GOAL:*** *To increase prodrug stability using masking groups that are not recognized by hydrolytic enzymes while being labile to Pd chemistry*



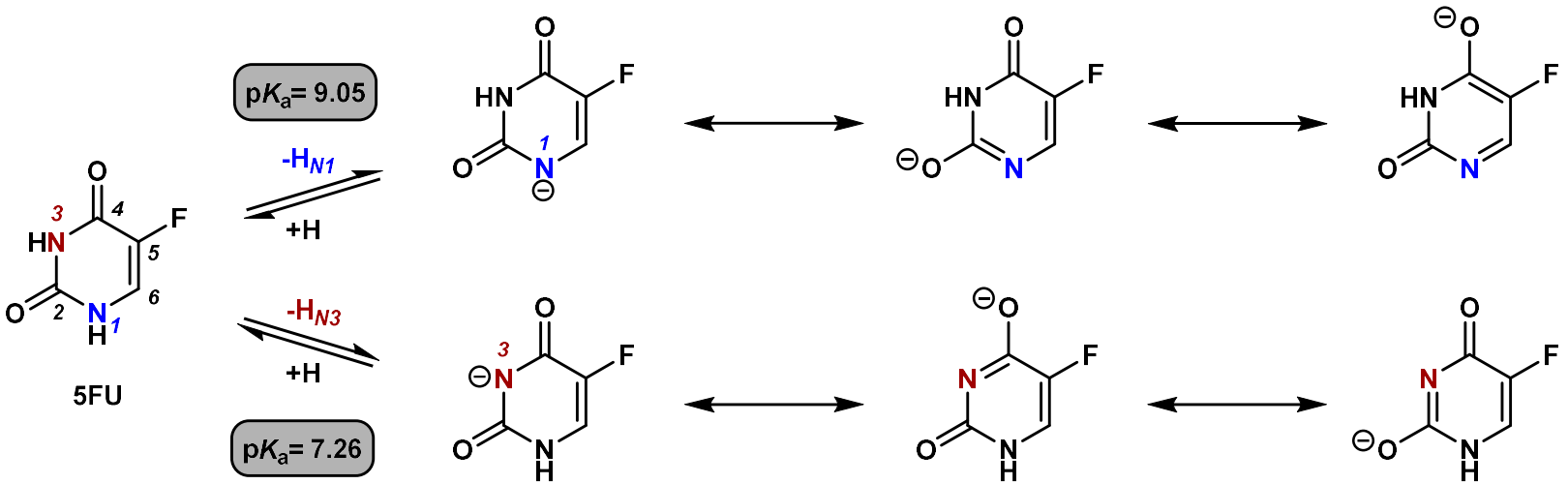

***DRUG's Mode of Action:***
*5FU is converted intracellularly into cytotoxic nucleotidic metabolites, which inhibit directly thymidylate synthase or incorporates into RNA and DNA to disrupt normal cell functions*

***CHEMICAL CHALLENGE!!*** *According to the literature, palladium-mediated N-dealkylations in water typically require temperatures incompatible with cell survival (>80 °C)*

*Nat Commun* **2014**, 5, 3277

# Drugs as “leaving groups”

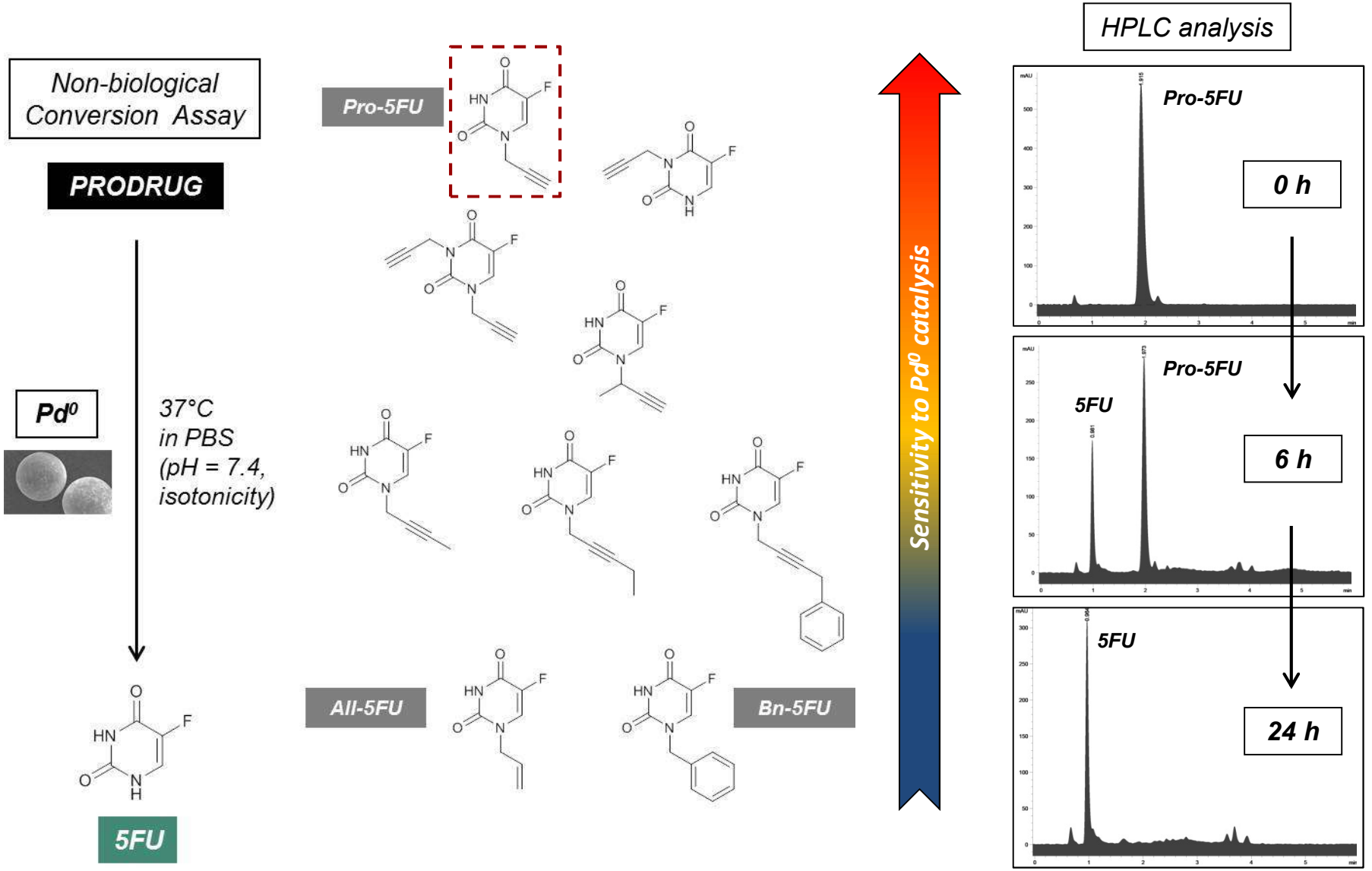
***CHEMICAL CHALLENGE!!*** *According to the literature, palladium-mediated N-dealkylations in water typically require temperatures incompatible with cell survival (>80 °C)*




***LACTAM LACTIM TAUTOMERY:*** *Endocyclic NH groups of 5FU possess relatively low $pK_a$ values due to its unique properties.*

*Nat Commun* **2014**, 5, 3277

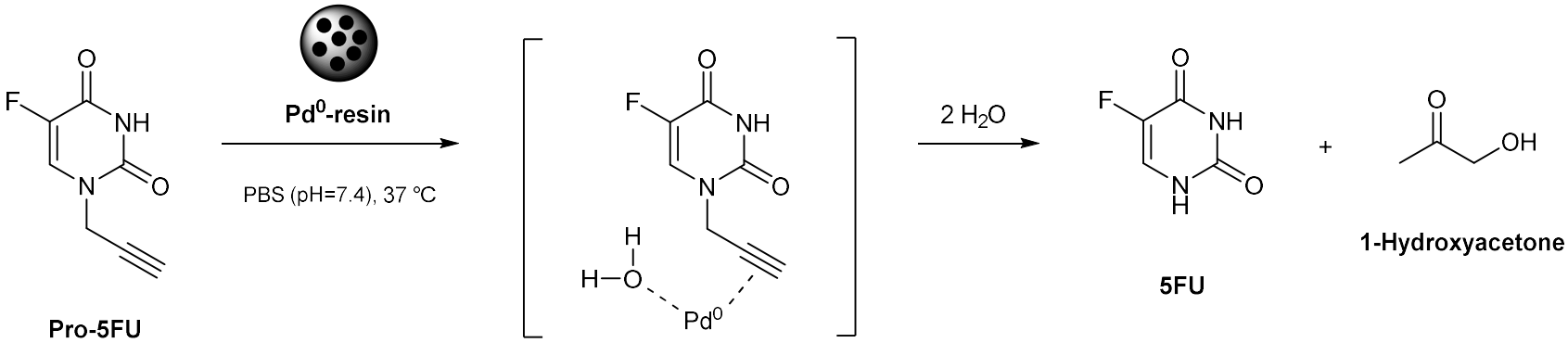
# Prodrug deprotection

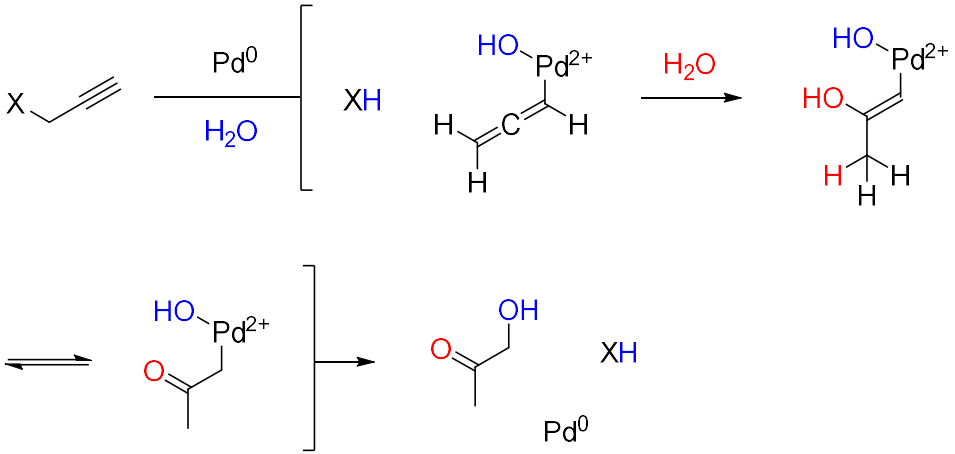


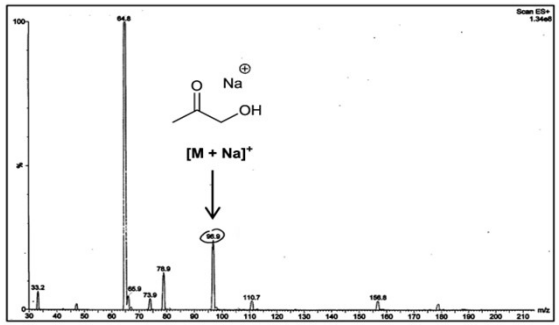

*Front Chem* **2014**, 2, 56 *Nat Commun* **2014**, 5, 3277

# Pd-mediated depropargylation

*Análisis HPLC*



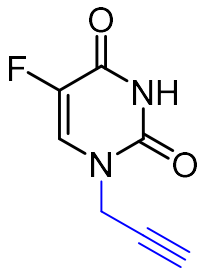




*J Med Chem* ***2014***, 57, 5395

# Prodrug Safety and Activation

*Alkylation of the N1 position of 5FU (cytotoxic drug used to treat colorectal and pancreatic cancer) resulted in biochemically-stable inactive derivatives (reduction of cytotoxicity >500 fold).*

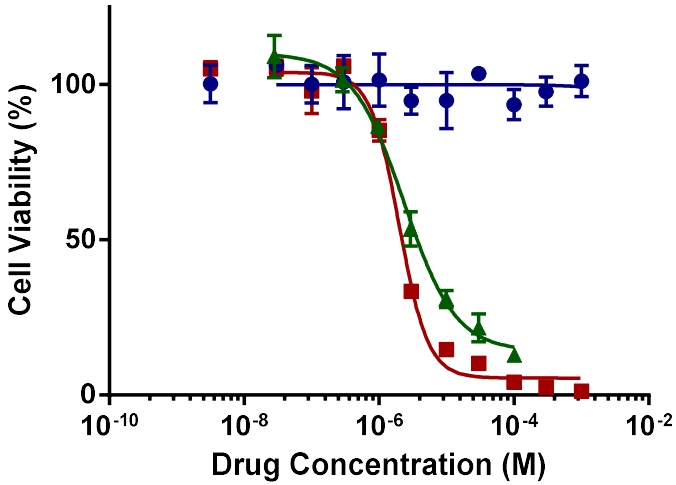
**Pro-5FU**




***COLORECTAL Cancer Cells***

| | $EC_{50}$ |
|---|---|
| Pro-5FU | >1 mM |
| 5FU | 2.0 uM |
| Pd + Pro-5FU | 2.4 uM |

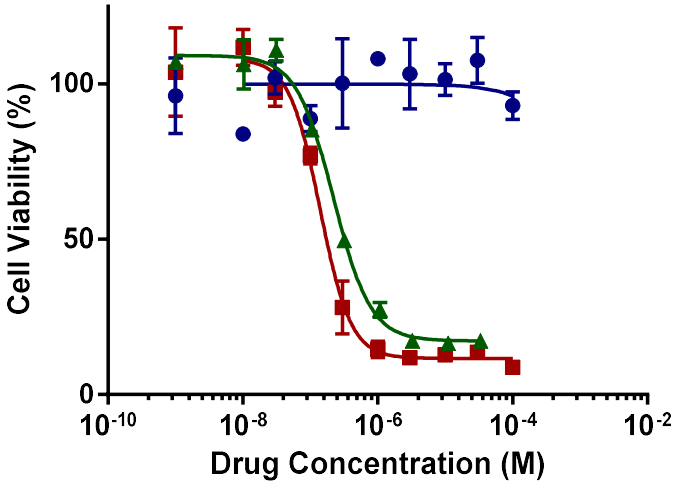
***> 500 fold***




***PANCREATIC Cancer Cells***

| | $EC_{50}$ |
|---|---|
| Pro-5FU | >100 uM |
| 5FU | 136 nM |
| Pd + Pro-5FU | 209 nM |

***> 700 fold***



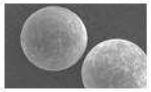

*Nat. Commun.* **2014**, 5, 3277

# Visualizing inactive prodrug and devices

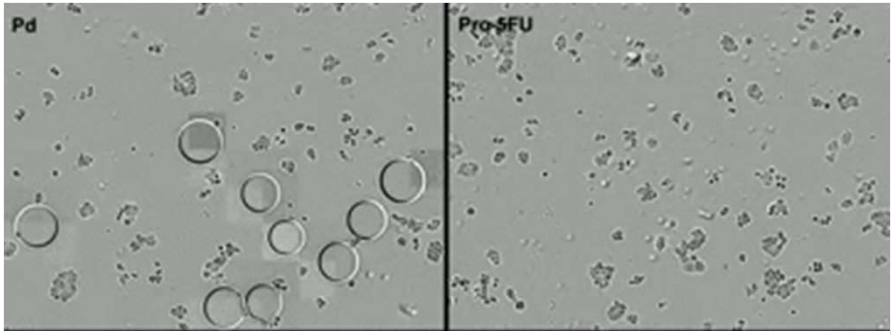
*Chemical masking of **5FU** (drug used to treat colon and pancreatic cancer) results in a completely inactive derivative (reduction of cytotoxicity >500 fold).*

**Palladium**

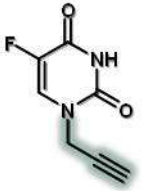
**SAFE**



$Pd^0$-resins




***Real-time visualization (5 days) of cell proliferation (colorectal cancer cells)***




*Pro-5FU*

**PRODRUG**

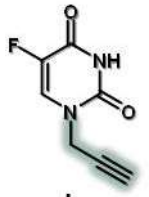
**SAFE**

*Nat. Commun.* **2014**, 5, 3277

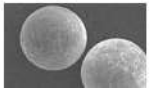
# Visualizing prodrug activation

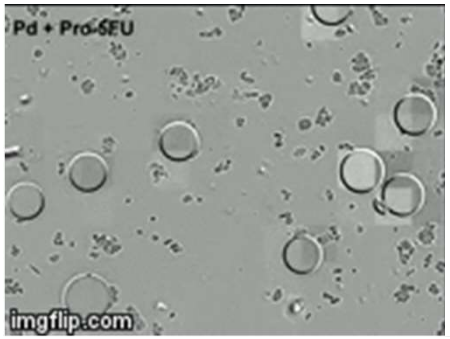
*Combination of inactive **Pro-5FU** and **Pd-devices** mediated strong cytotoxic activity, equivalent to that of **5FU**, demonstrating the in situ manufactured of the drug*

**PRODRUG**
**+**
**Palladium**




+



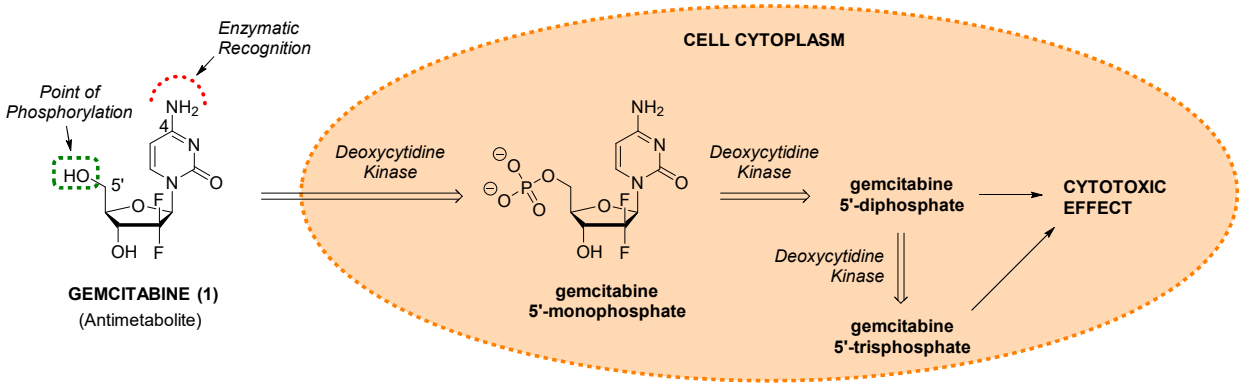



**CYTOTOXIC**

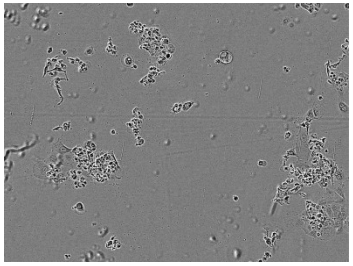
***Real-time visualization (5 days) of cell proliferation (colorectal cancer cells)***

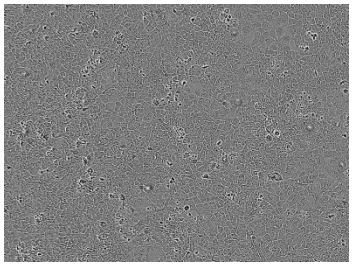
*Nat. Commun.* **2014**, 5, 3277

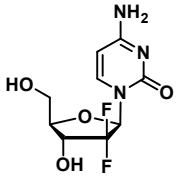
# $Pd^0$-Labile Gemcitabine Prodrugs

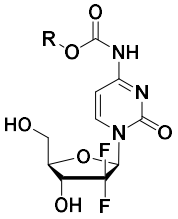


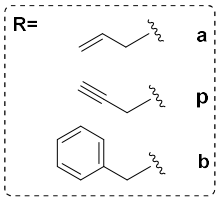

***Gemcitabine (Gemzar):***
*antimetabolite >> 1st line therapy against pancreatic cancer*
***Masking Strategy:***
*5'-OH of ribose and 4-amino group of the cytosine base*





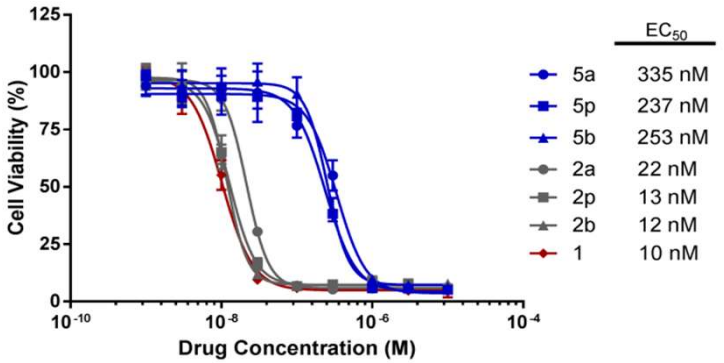

Gemcitabine (1)


5a,p,b




## Bioorthogonality Study

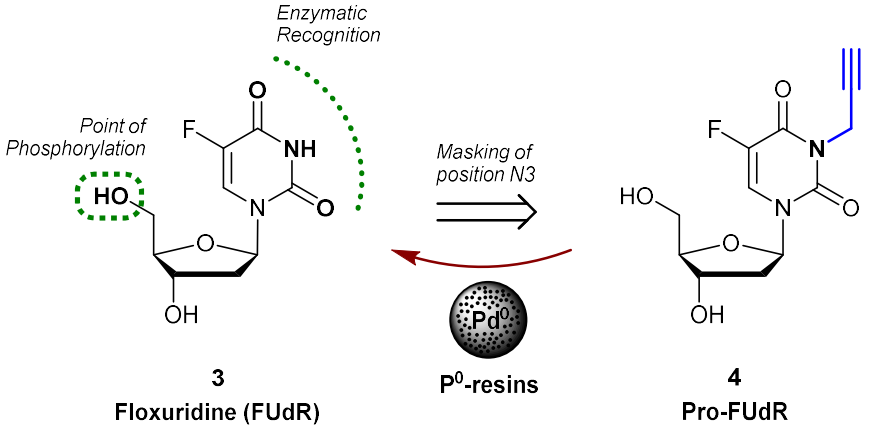
***Drug vs Prodrugs= x24-33 difference in activity***




*J Med Chem* ***2014***
doi: 10.1021/jm500531z

# $Pd^0$-Labile Floxuridine Prodrugs

***Floxuridine (FUdR):***
*antimetabolite >> treatment of colorectal cancer*
***Masking Strategy:***
*3-NH group of the 5-fluorouracil base*

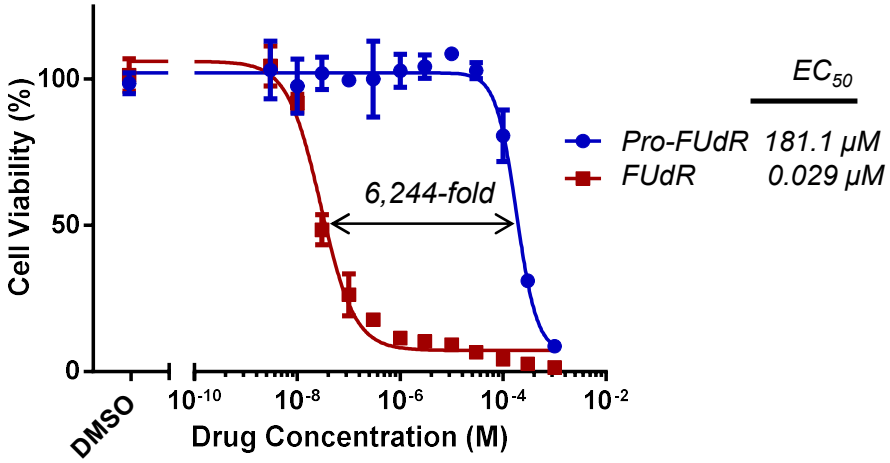


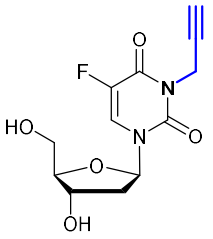

**3**
**Floxuridine (FUdR)**

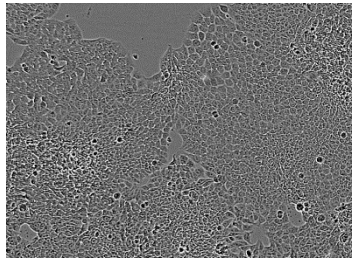
**4**
**Pro-FUdR**

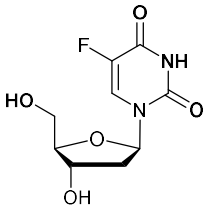
## *Bioorthogonality Study*

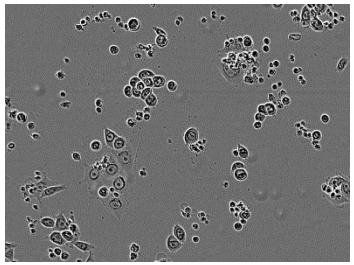
***Drug vs Prodrug= x6,000 difference in activity***



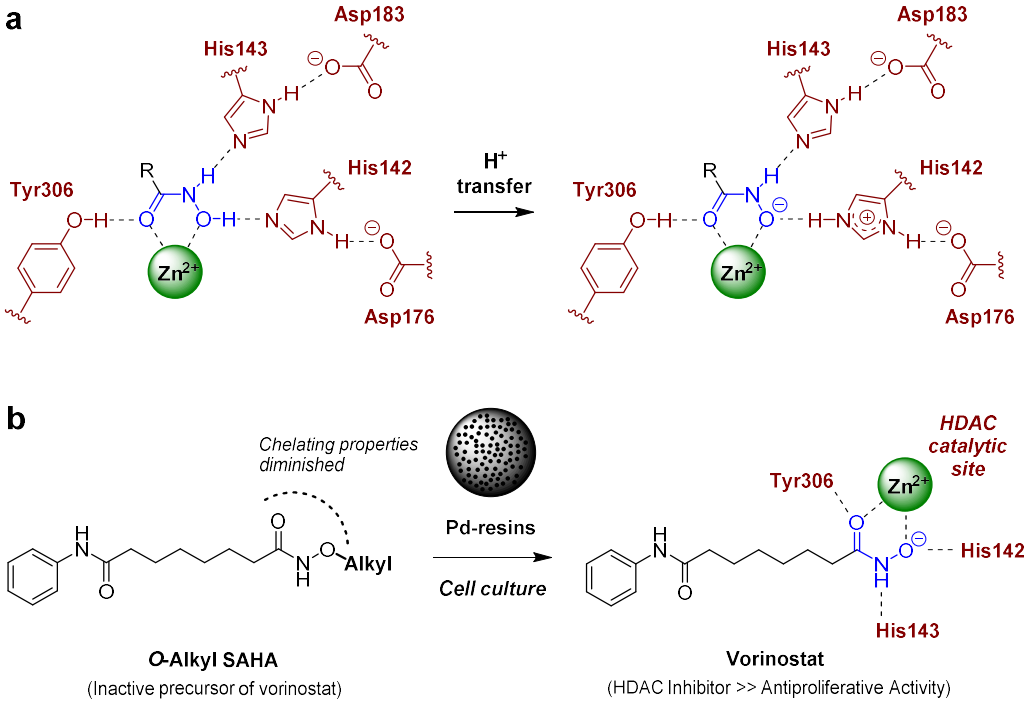



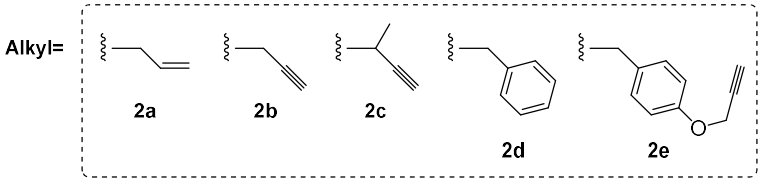


*Sci. Rep.* **2015**, *5*, 9329

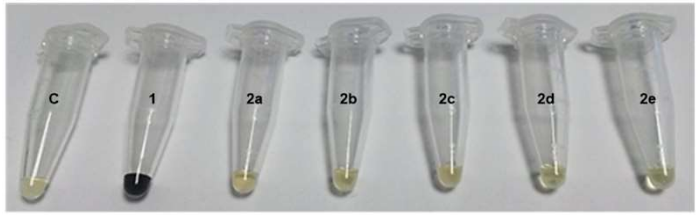
# Bioorthogonal release of hydroxamic acids




***Vorinostat*** *is an anticancer agent that targets a group of enzymes called histone deacetylases. This drug possesses an hydroxamic group that coordinates with a* $Zn^{2+}$ *atom at the enzyme's active site.*

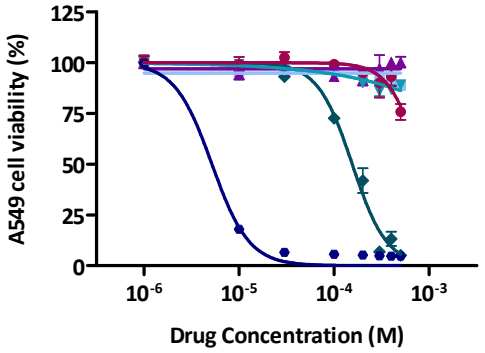
***Hypothesis****: Blockade of such group could reduce the capacity of the chemical to chelate Zn cations at the active site of the enzyme, thus reducing its pharmacological activity*



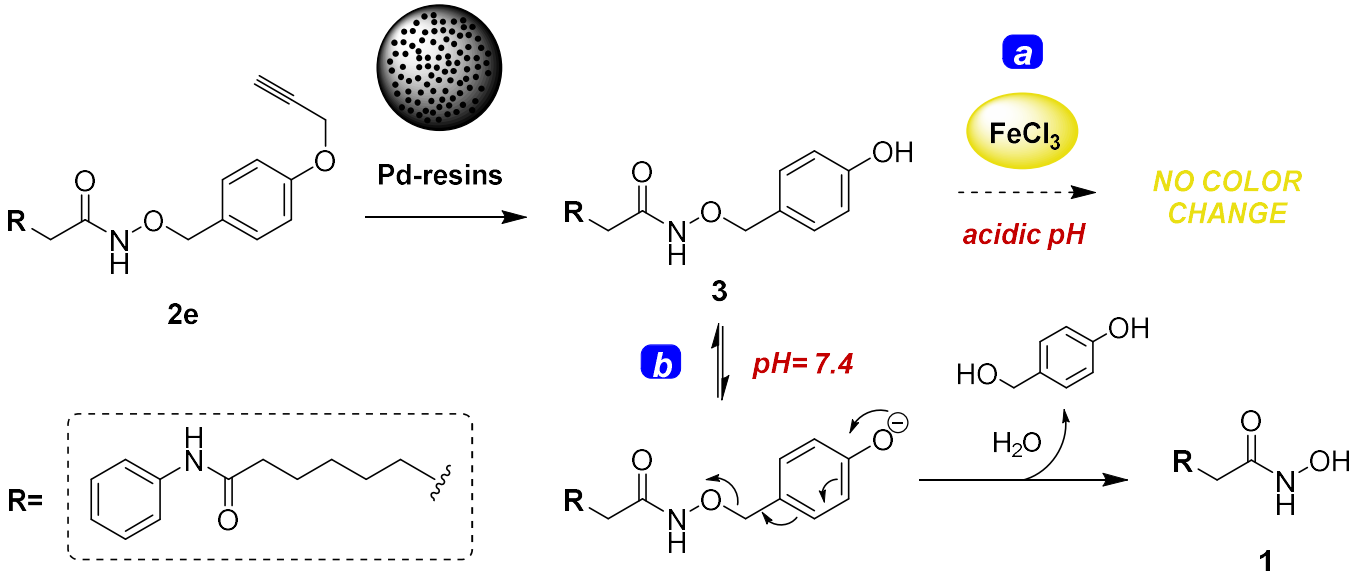

*J. Med. Chem.* **2016**, *59*, 9974

# Bioorthogonal release of hydroxamic acids

*O-alkylated derivatives of vorinostat exhibit low / null anticancer activity in lung cancer cells*



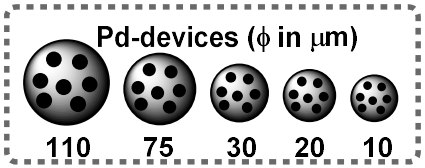

| | $EC_{50}$ A549 | $EC_{50}$ U87G |
|---|---|---|
| 1 | 4.8 µM | 11.0 µM |
| 2a | > 500 µM | > 500 µM |
| 2b | 155.0 µM | 283.8 µM |
| 2c | > 500 µM | > 500 µM |
| 2d | > 500 µM | > 500 µM |
| 2e | > 500 µM | > 500 µM |

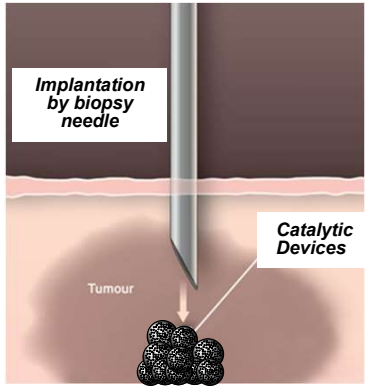



*J. Med. Chem.* **2016**, *59*, 9974

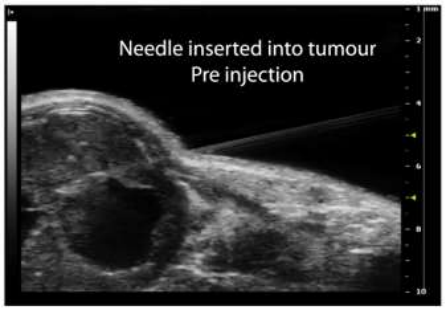
# Platform Technology

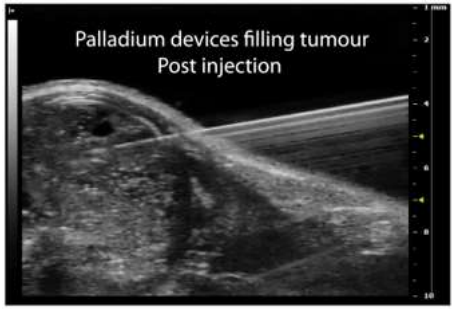
| Catalyst | | |
|---|---|---|
| Palladium | Safe*<br>**Classified as Biocompatible metals* | 16w in mice<br>84d in rats |
| Gold | | |



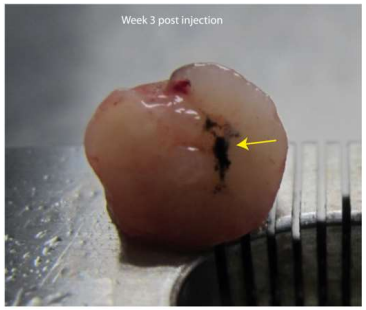

| Chemotherapy | Main Cancer Indication |
|---|---|
| 5FU | Colorectal, Breast, Pancreatic, Oesophageal, Vaginal, Cervical, Anal |
| Irinotecan | Colorectal, Lung, Pancreatic, Head & Neck |
| Doxorubicin | Breast, Pancreatic, GI, Bladder, kidney, Bone, CNS, Colorectal |



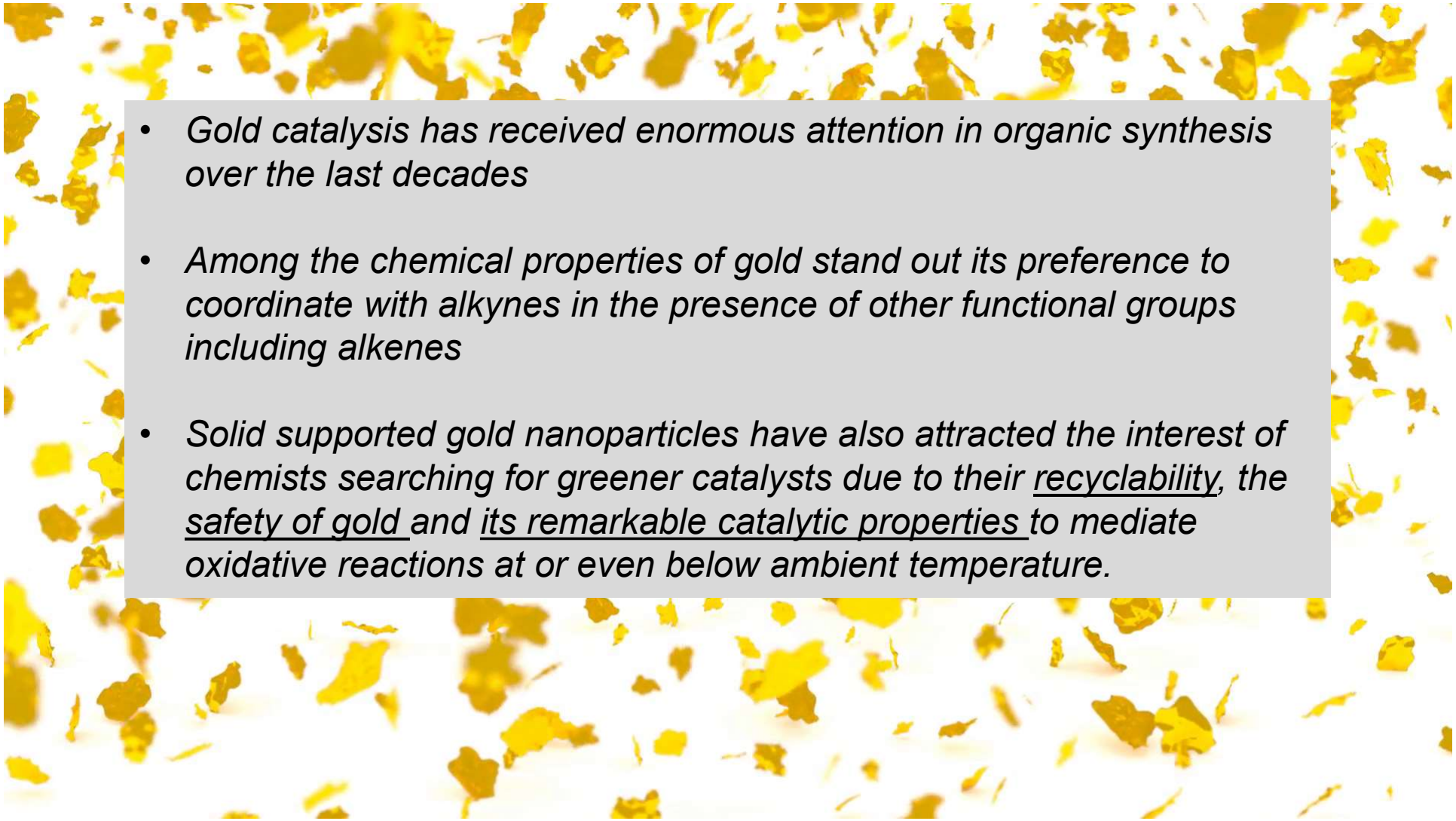

Devices are *echogenic* >> intratumoural insertion guided by ultrasound imaging

Devices do not “move” from the point of injection




*Chem. Sci.* **2018**, *9*, 7354-7361

# Beyond Palladium



- *Gold catalysis has received enormous attention in organic synthesis over the last decades*
- *Among the chemical properties of gold stand out its preference to coordinate with alkynes in the presence of other functional groups including alkenes*
- *Solid supported gold nanoparticles have also attracted the interest of chemists searching for greener catalysts due to their recyclability, the safety of gold and its remarkable catalytic properties to mediate oxidative reactions at or even below ambient temperature.*

# Our work in the News

DAILY EXPRESS

## Role found for gold in cancer fight

By Katrine Bussey

TINY flecks of gold could be used in the fight against cancer, new research has suggested.

Scientists at Edinburgh University have just completed a study which shows the precious metal increased the effectiveness of drugs used to treat lung cancer cells.

Minute fragments, known as gold nanoparticles, were encased in a chemical device by the research team.

While this has not yet been tested on humans, it is hoped it could one day be used to reduce side-effects of current chemotherapy treatments by precisely targeting diseased cells without damaging healthy tissue.

Gold is a safe element and has the ability to accelerate chemical reactions.

Researchers at the university discovered properties of the metal that allow these catalytic abilities to be accessed in living things without any side-effects.

The device was shown to be effective after being implanted in the brain of a zebrafish, suggesting it can be used in living animals.

The study was carried out in collaboration with researchers at the University of Zaragoza's Institute of Nanoscience of Aragon in Spain, with funding coming from Cancer Research UK (CRUK), and the Engineering and Physical Sciences Research Council.

Dr Asier Unciti-Broceta, from the University of Edinburgh's CRUK

BBC

Your account | News | Sport | Weather | iPlayer | TV | Radio

NEWS

Home | UK | World | Business | Politics | Tech | Science | Health | Education | Entertainment

Scotland | Scotland Politics | Scotland Business | Edinburgh, Fife & East | Glasgow & West

### Gold 'could be used in cancer treatment'

7 August 2017 | Edinburgh, Fife & East Scotland | Share


**Tiny flecks of gold could be used in the fight against cancer, new research has suggested.**

Scientists at Edinburgh University found the precious metal increased the effectiveness of drugs used to treat lung cancer cells.

Minute fragments, known as gold nanoparticles, were encased in a chemical by the research team.

The research involved zebrafish but the team are hopeful the technique could be used to develop human treatments.

Gold is a safe element which can accelerate - or catalyse - chemical reactions.

Daily Scottish Mail

## The new weapon in the fight with cancer... gold

### Study shows tiny flecks of metal can boost effectiveness of drugs

By Daily Mail Reporter

TINY flecks of gold could be implanted in humans to combat cancer, new research has suggested.

Scientists at Edinburgh University have completed a study which shows the precious metal increased the effectiveness of drugs used to treat lung cancer cells.

The team encased minute fragments, known as gold nanoparticles, in a prototype chemical

"There is still work to do before we can use this on patients, but this study is a step forward. We hope that a similar device in humans could one day be implanted by surgeons to activate chemotherapy directly in tumours and reduce harmful effects to healthy organs."

Dr Áine McCarthy, Cancer Research UK's senior science information officer, said: 'By developing new, better ways of delivering cancer drugs, studies like this have the potential to improve cancer treatment and reduce side effects. In particular, it could help improve treatment for brain tumours and other hard-to-treat cancers.

'The next steps will be to see if this method is safe to use in people, what its long and short-term side effects are, and if it's a better way to treat some cancers.'

The research has been published in the scientific journal Angewandte Chemie.

Gold already has a variety of medical uses as it is highly resistant to bacteria. It is used in wires for pacemakers and in stents which support weak blood vessels in the treatment of heart disease. Scientists frequently used the freshwater zebrafish in cancer research for a number of reasons.

Their translucent skin allows researchers to observe tumour growths and reductions without having to use medical scanners.

They also reproduce at an incredibly high rate. A breeding pair can produce up to 300 embryos a week, allowing scientists to conduct thousands of tests at a time to ensure the reliability of their results.

Though only 2.5in in length they also share a number of genes with humans, meaning they can develop most of the tumours we can. A staggering 84 per cent of genes linked with human disease have a zebrafish counterpart.

### 'Reduce harmful effects to organs'

The News Matrix

The day at a glance

SCIENCE

**Gold could help to fight lung cancer**

Gold could soon be used to help treat cancer, research has suggested. Scientists used tiny fragments of the precious metal to target lung cancer cells. Dr Asier Unciti-Broceta, from Edinburgh University, said the metal could be used to release drugs inside tumours "very safely". PAGE12

SPUTNIK

### This Is a Big Thing': Gold Nanoparticles Could Be Used to Treat Cancer

**Tiny flecks of gold could be used in the fight against cancer, new research has suggested. Scientists at Edinburgh University have completed a study which shows the precious metal increased the effectiveness of drugs used to treat lung cancer cells.**

Researchers, led by Dr. Asier Unciti-Broceta, have discovered the gold has the ability to act as a catalyst for chemotherapy.

Christine CRUK @CRUKEastScot

Exciting study from @EdinORC Dr Asier Unciti-Broceta - using gold to increase the effectiveness of cancer treatments goo.gl/u5x8kc

10:13 AM - Aug 7, 2017

NEWS

LATEST | MOST READ | MOST DISCUSSED

Outgoing US Envoy to Russia Confesses Sanctions on Russia 'Blunt Instrument'

New Power Balance: Will Turkey 'Play a Key Role' in Change of World Order?

Car Bomb Kills 12 People, Injures 24 in Baghdad - Reports

Trump Not to Visit Houston, Texas Amid Massive Flooding Over Hurricane Harvey

Black-Clad Antifa Attack Peaceful Berkeley Rally, Ignite Furor on Social Media

All news

RECOMMENDED

Tom Hank's Net Worth Left Us

METRO

## Gold could aid cancer treatment

**TINY flecks of gold could be used in the fight against cancer, new research has suggested.**

**Scientists at Edinburgh University have completed a study which shows the precious metal increased the effectiveness of drugs used to treat lung cancer cells.**

**Minute fragments, known as gold nanoparticles, were encased in a chemical device. While this has not yet been tested on humans, it is hoped such a device could one day be used to reduce side effects of current chemotherapy treatments by precisely targeting diseased cells without damaging healthy tissue.**

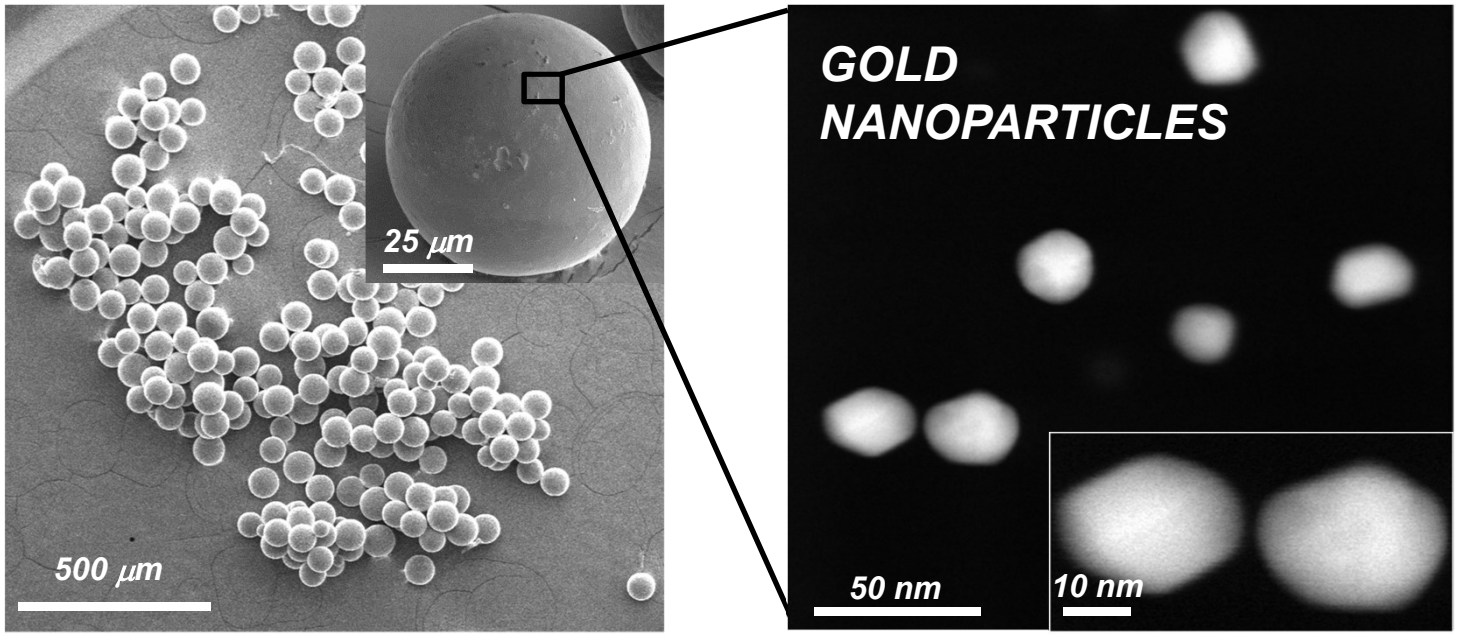
**The device was shown to be effective after being implanted in the brain of a zebrafish, suggesting it can be used in living animals.**

**The study was carried out in collaboration with the University of Zaragoza, with funding coming from Cancer Research UK and the Engineering and Physical Sciences Research Council.**

# Gold-Functionalized Devices

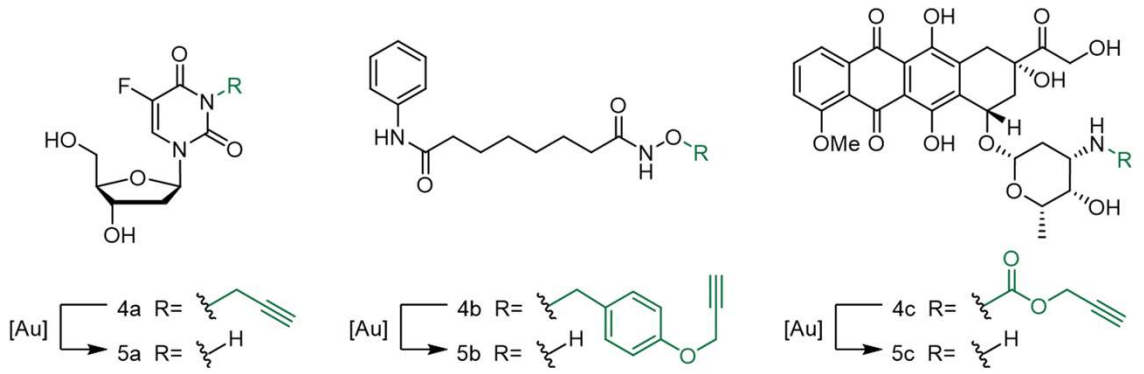
*Because of the high affinity of thiols for gold and their ubiquitous presence in peptides and proteins, the attractive catalytic properties of gold nanoparticles have not been accessible in living environments.*

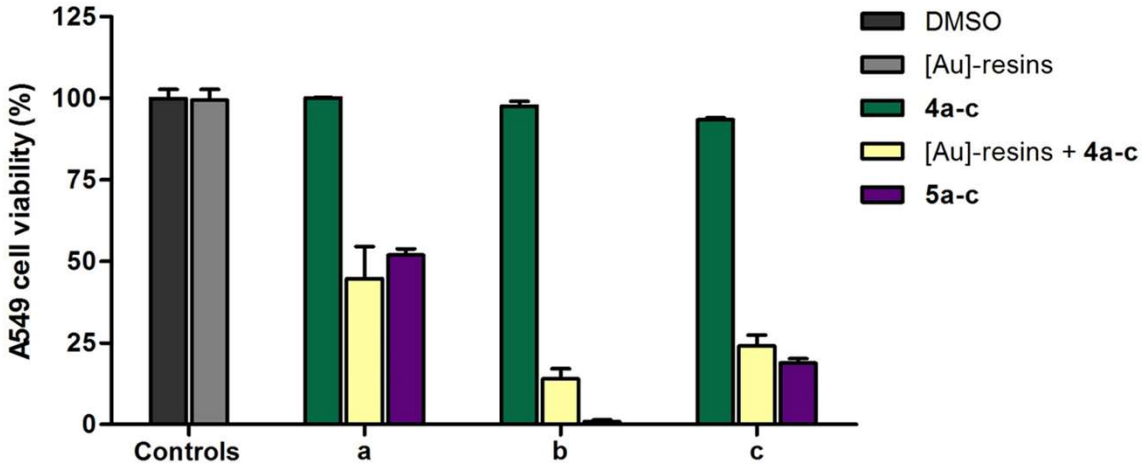
*We envisioned that embedding Au-NP in a solid support would serve to protect the metal nanostructures from large thiol-rich biomolecules, while allowing the free entry of alkyne-functionalized small molecules to undergo gold-mediated chemistries even in biological systems.*




*Angew. Chemie* **2017**, 10.1002/anie.201705609

# Prodrug Activation Studies

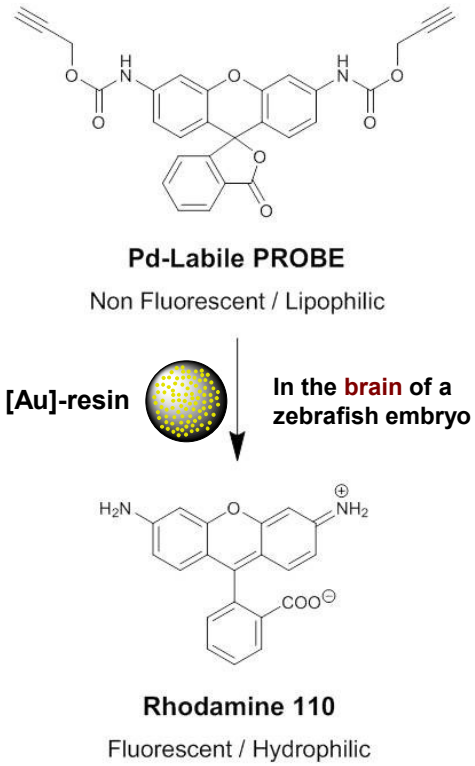


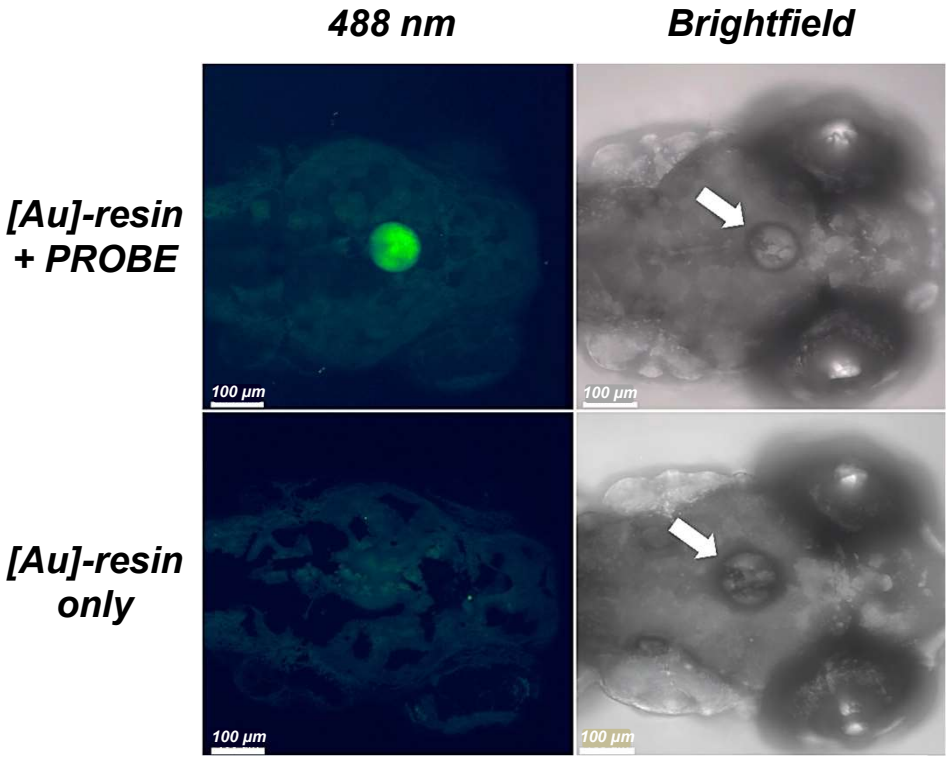




*As expected prodrugs **4a-c** did not elicit any effect on their own. On the contrary, potent anticancer activity was displayed in combination with [Au]-resins, unequivocal evidence that the active drugs are released in situ by heterogeneous gold chemistry.*

# Bioorthogonal chemistry in the brain

*First example of heterogeneous metal-catalyzed release of a chemical reporter performed in the brain of a living animal*

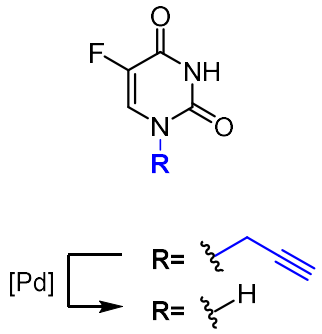


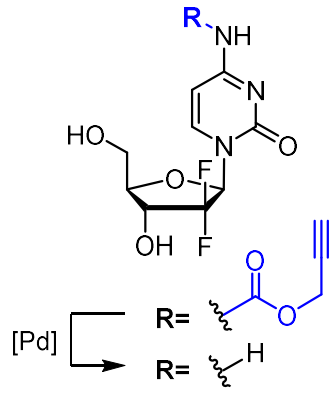


*Angew. Chemie* **2017**, *56*, 12548

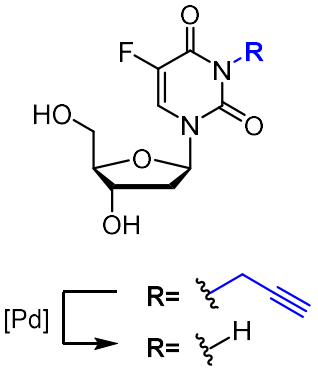
# Palladium activated prodrugs



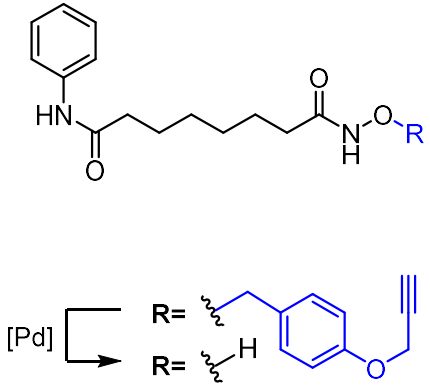

*Nat. Commun.* **2014**, *5*, 3277



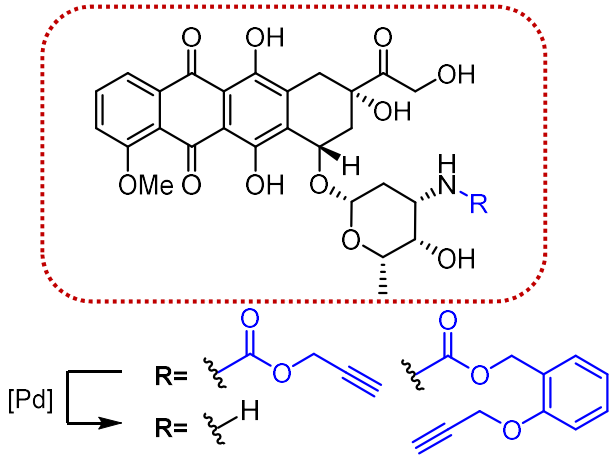

*J. Med. Chem.* **2014**, *57*, 5395




*Sci. Rep.* **2015**, *5*, 9329

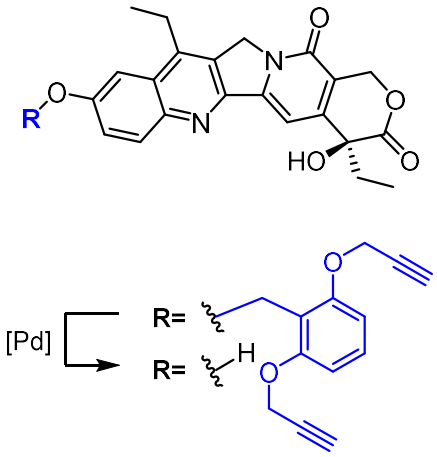



*J. Med. Chem.* **2016**, *59*, 9974




*Angew. Chemie* **2017**, *56*, 12548

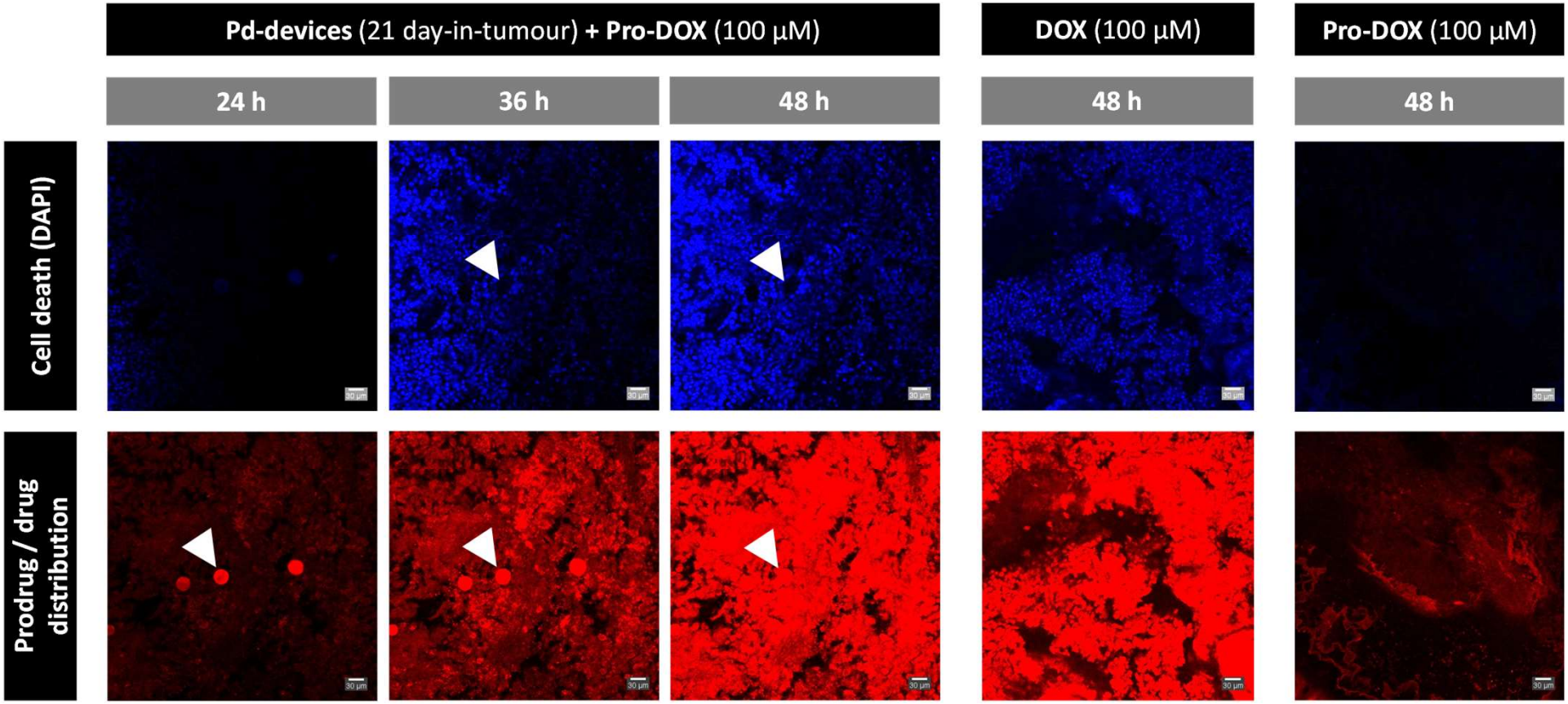
*Chem. Sci.* **2018**, *9*, 7354-7361




*Chem. Eur. J.* **2018**, *24*, 16783-16790

***PATENTED***

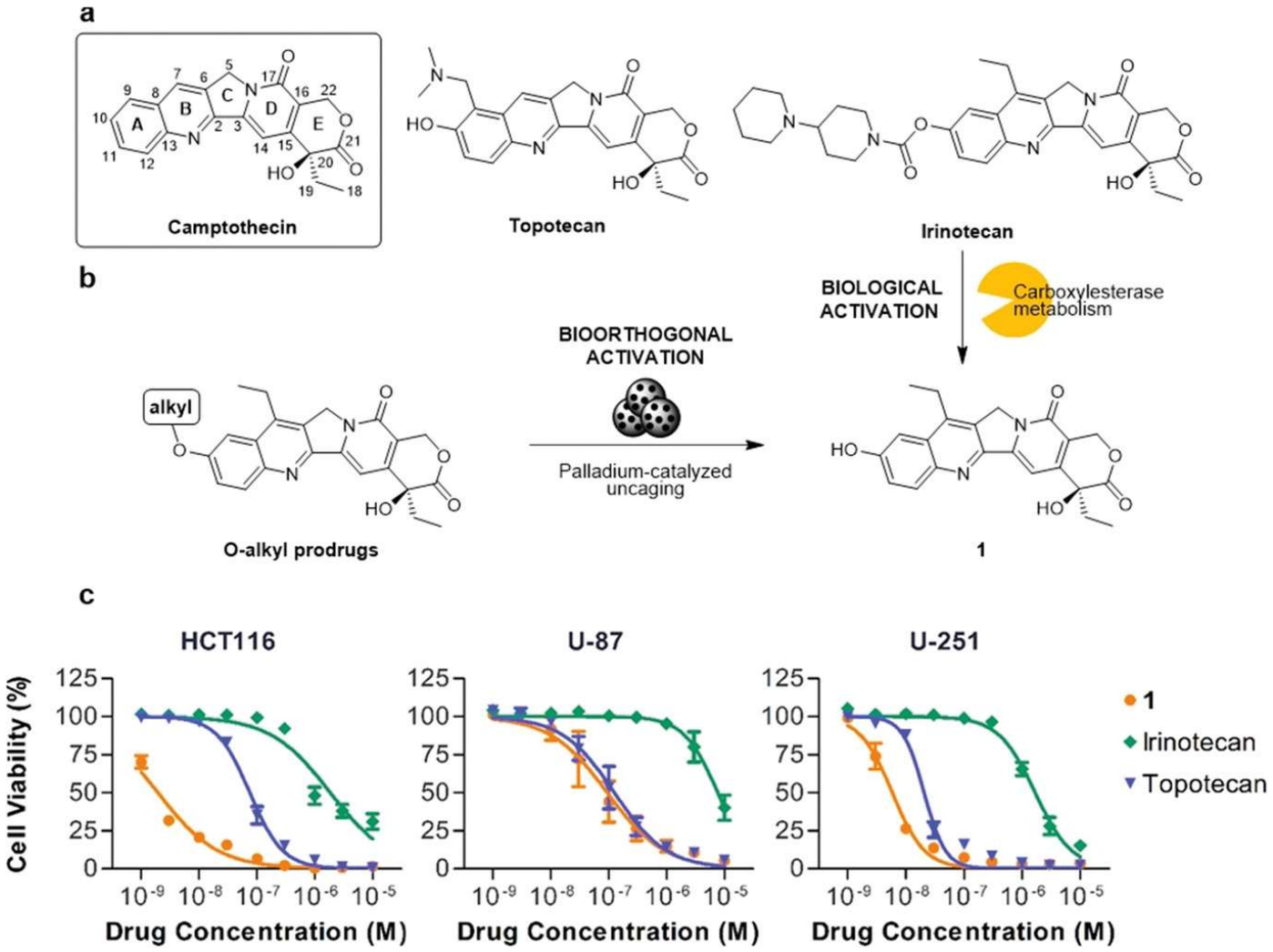
# *Ex vivo* activation of chemotherapy




***Ex vivo* Pd-mediated release of DOX from an inactive precursors in a prostate tumour explant**

*Chem. Sci.* **2018**, *9*, 7354-7361

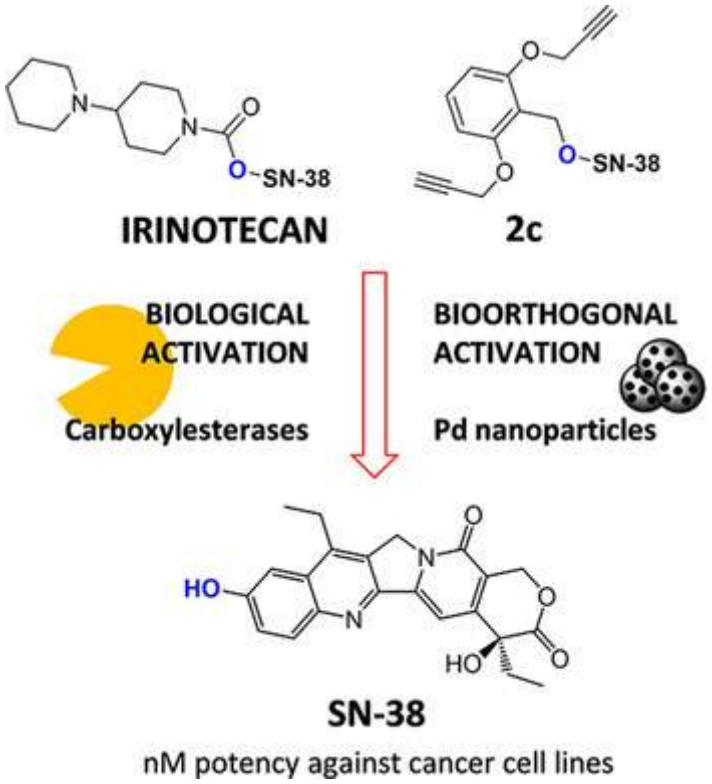
# Bioorthogonal Uncaging of SN-38

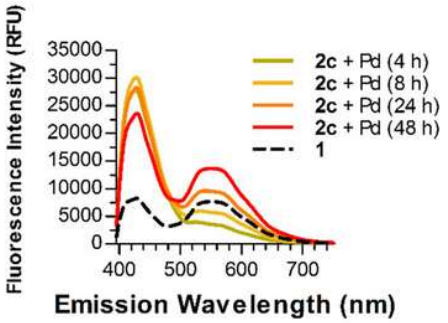


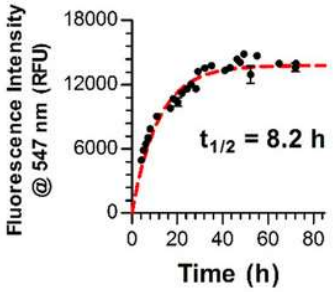

*Chem. Eur. J.* **2018**, *24*, 16783-16790

# Bioorthogonal Uncaging of SN-38

*Palladium-functionalized microdevices were used to uncage newly developed prodrug that releases SN-38 (irinotecan's active metabolite) in combination with previously known anticancer prodrug of 5FU.*



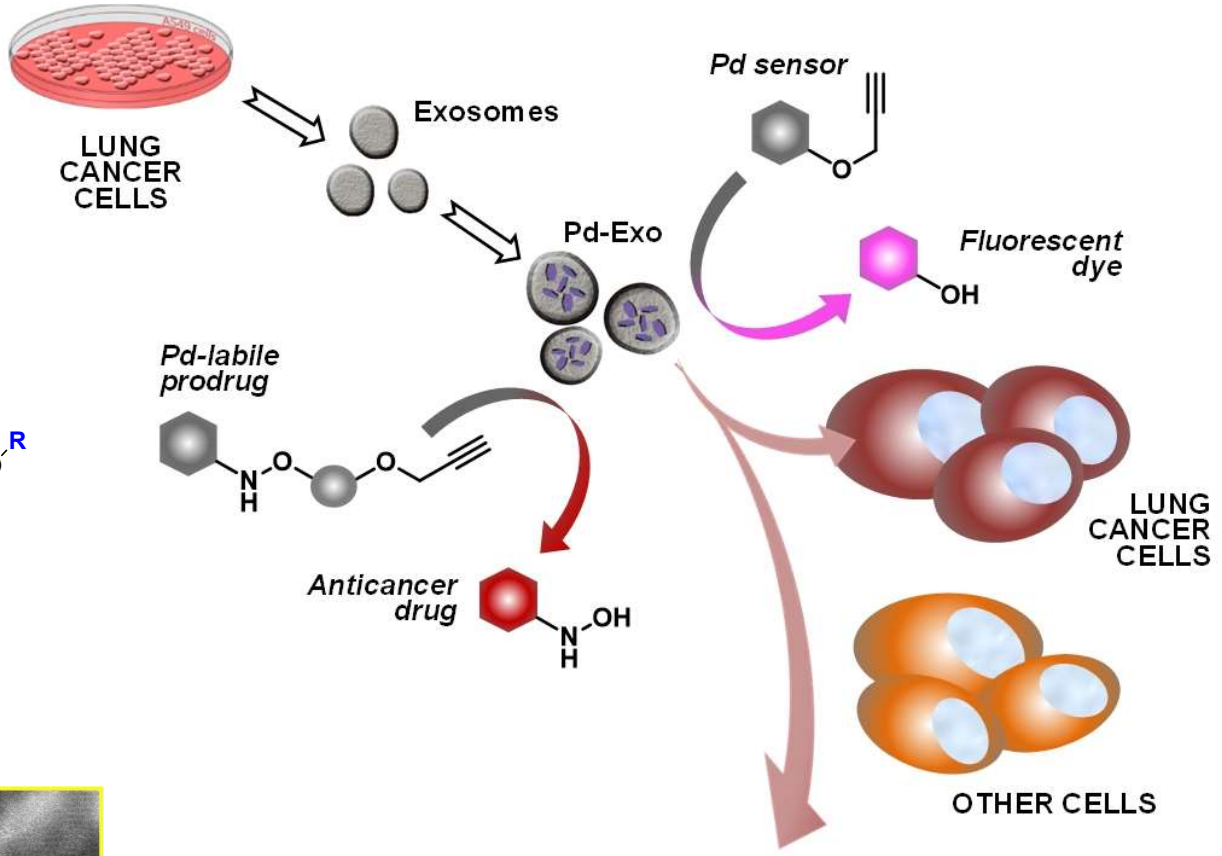



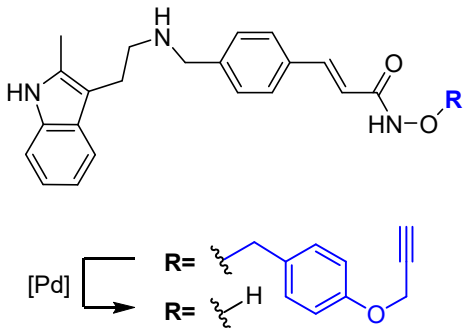



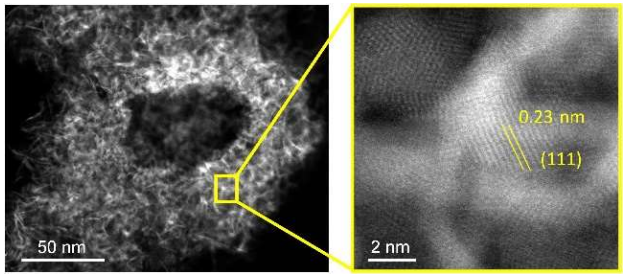

*Chem. Eur. J.* **2018**, *24*, 16783-16790

# Targeted Bioorthogonal Catalysis

*Cancer-derived exosomes loaded with ultrathin Palladium nanosheets*




*Exosome-directed catalyst prodrug therapy*

*Nat. Catalysis* **2019**, 10.1038/s41929-019-0333-4