by an E3 ubiquitin ligase that binds to a protein and adds ubiquitin molecules to the protein. The E3 ubiquitin ligase is part of a pathway that includes E1 and E2 ubiquitin ligases, which make ubiquitin available to the E3 ubiquitin ligase to add to the protein.

To harness this degradation pathway, molecular constructs known as chemical inducers of degradation (CIDEs) bring together an E3 ubiquitin ligase with a protein that is to be targeted for degradation. To facilitate a protein for degradation by the proteasome, the CIDE is comprised of a group that binds to an E3 ubiquitin ligase and a group that binds to the protein target for degradation. These groups are typically connected with a linker. This CIDE can bring the E3 ubiquitin ligase in proximity with the protein so that it is ubiquitinated and marked for degradation. However, the relatively large size of the CIDE can be problematic for targeted delivery, as well as contribute to undesirable properties, such as fast metabolism/clearance, short half-life, and low bioavailability.

There is an ongoing need in the art for improving CIDEs, including enhancing targeted delivery of CIDEs to cells that contain the protein target. The subject matter described herein addresses this and other shortcomings in the art.

BRIEF SUMMARY

In one example, the subject matter described herein is directed to conjugated or covalently linked Ab-CIDEs, wherein the positions of the covalent bonds that connect the components of the Ab-CIDE: Antibody (Ab), Linker 1 (L1), Linker 2 (L2), protein binding group (PB) and the E3 ligase binding group (E3LB), can be tailored as desired to prepare Ab-CIDEs having desirable properties, such as potency, in vivo pharmacokinetics, stability and solubility.

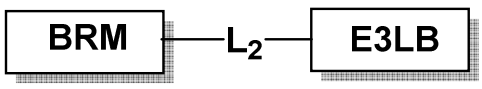
In one example, the subject matter described herein is directed to an Ab-CIDE having the chemical structure:

$$Ab\text{-}(L1\text{-}D)_p,$$

wherein,

Ab is an antibody;

D is a CIDE, or prodrug thereof, having the structure:



wherein,

BRM is a residue of a BRM-binding compound,

E3LB is a residue of an E3 ligase-binding compound, and
L2 is a moiety covalently linking BRM with E3LB;
L1 is a linker-1 covalently linking Ab to one of BRM, E3LB or L2; and
p is 1 to 16.

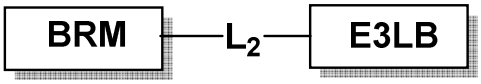
In one aspect, the disclosure provides a conjugate having the structure:

$$Ab\text{-}(L1\text{-}D)_p,$$

wherein,

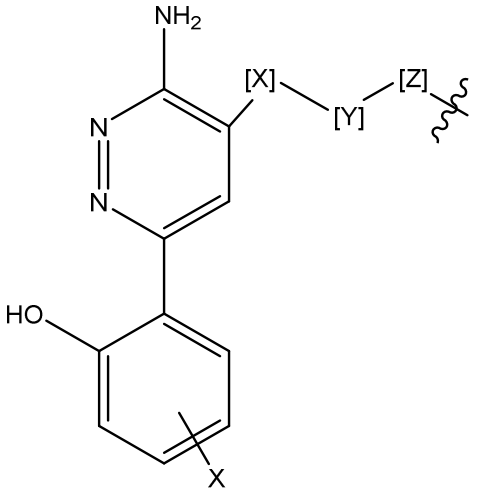
Ab is an antibody;

D is a CIDE, or prodrug thereof, the CIDE having the structure:

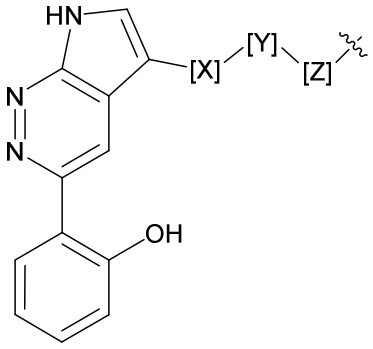


wherein,

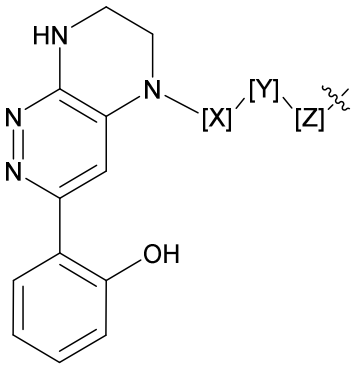
BRM is a residue of a BRM-binding compound, wherein the residue of a BRM-binding compound is a compound of formula (I-A), formula (I-B), formula (I-C), formula (I-D), or formula (I-E):



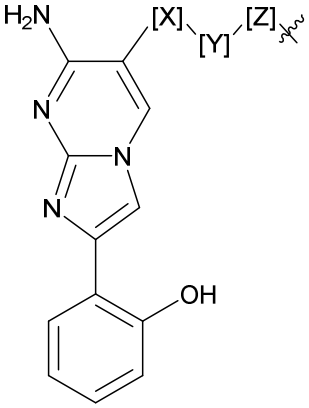
(I-A),



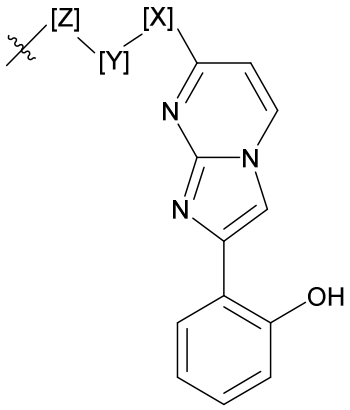
(I-B),



(I-C),



(I-D), wherein X is hydrogen or halogen; or



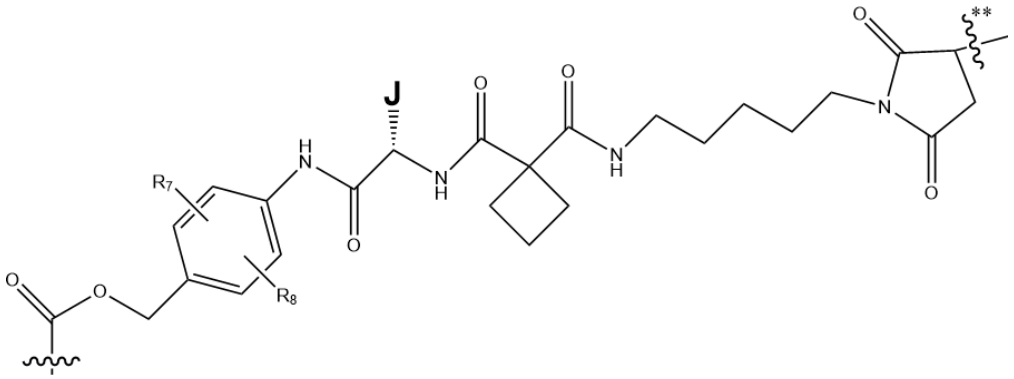
(I-E);

E3LB is a residue of an E3 ligase-binding compound, and

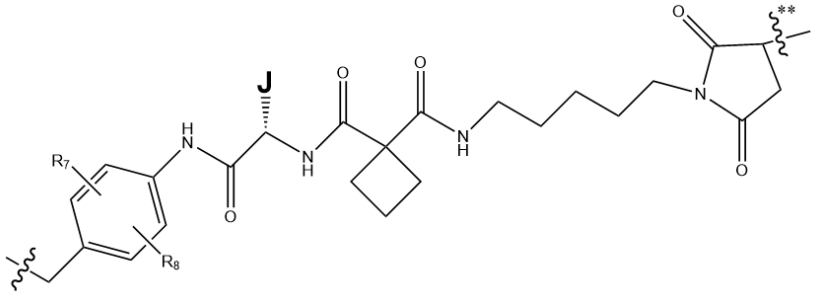
L2 is a linker covalently linking BRM and E3LB;

L1 is a linker covalently linking Ab to BRM, E3LB or L2;

wherein L1 is selected from the group consisting of:



, and



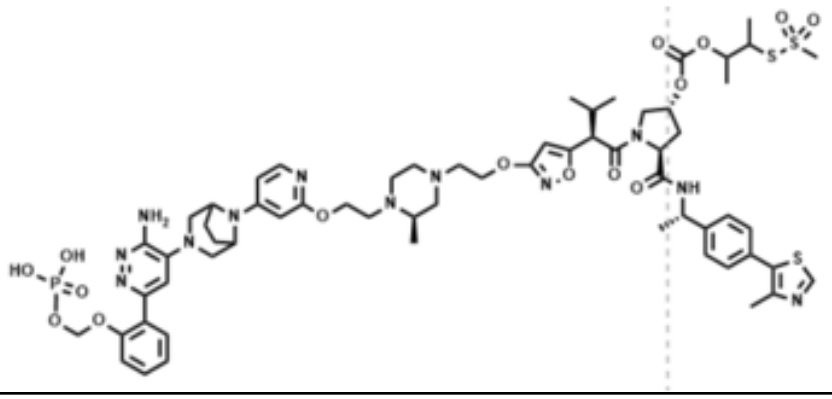
wherein,

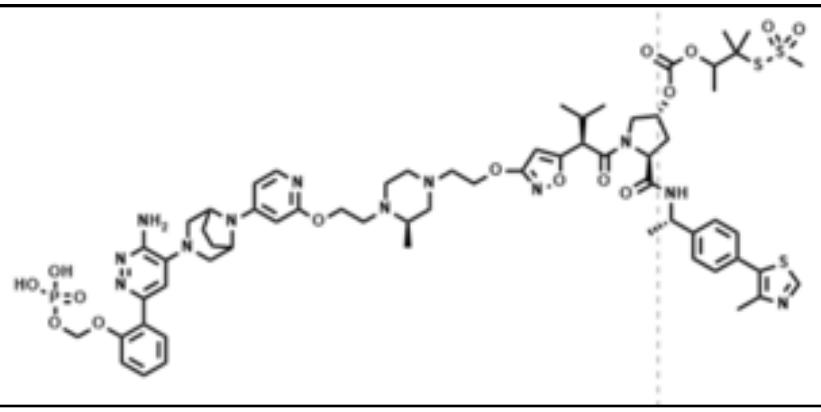
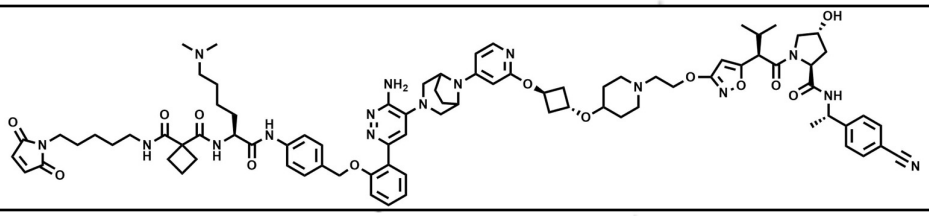
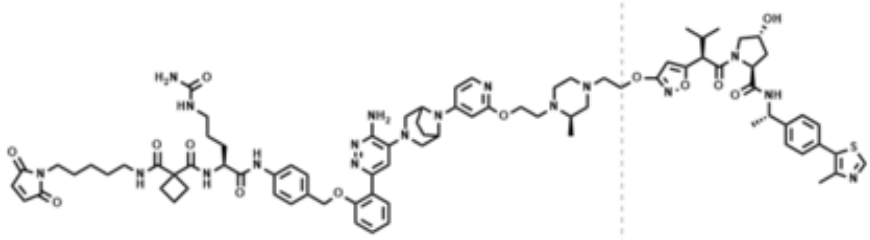
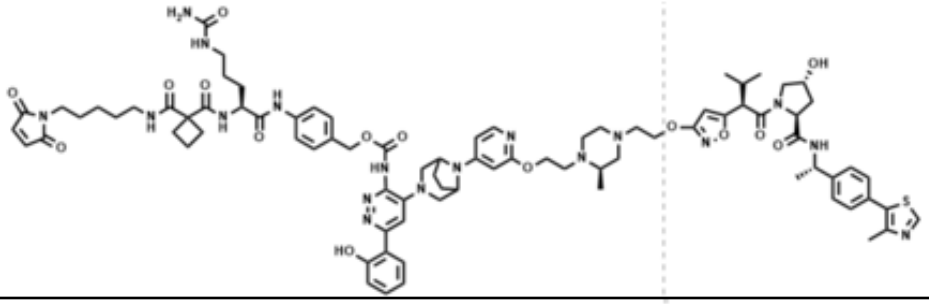
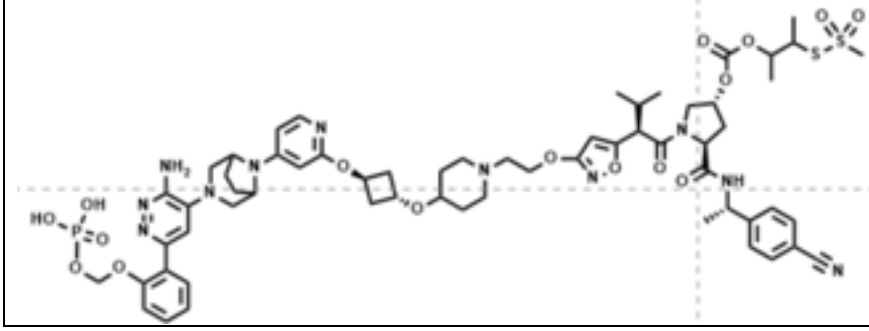
J is —CH_2-CH_2-CH_2-NH-C(O)-NH_2; —CH_2-CH_2-CH_2-CH_2-NH_2; —CH_2-CH_2-CH_2-CH_2-NH-CH_3; or —CH_2-CH_2-CH_2-CH_2-N(CH_3)$_2$; and

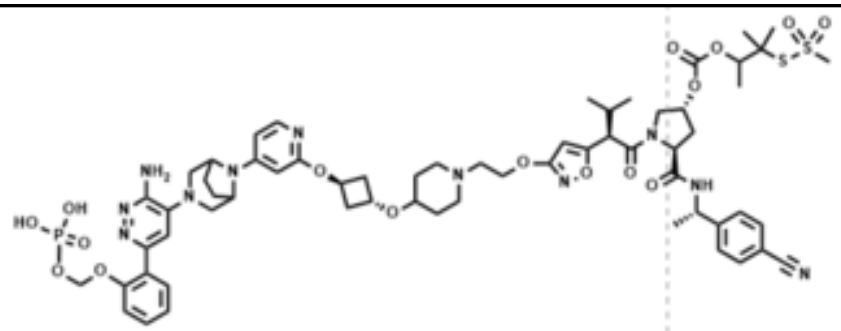
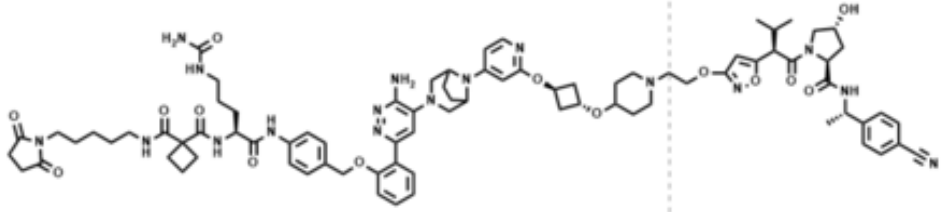
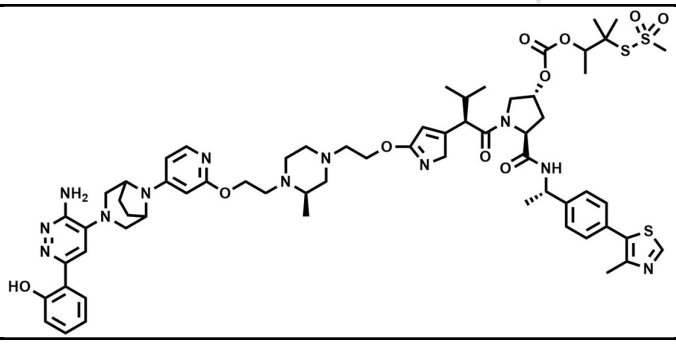
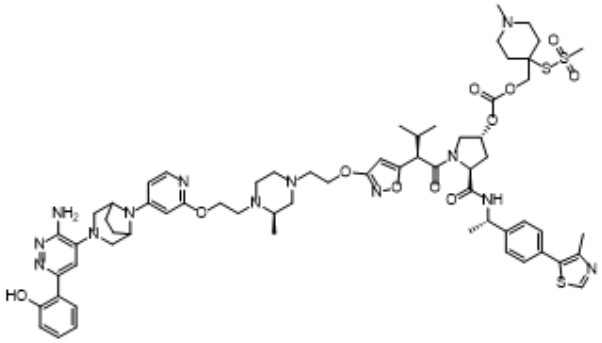
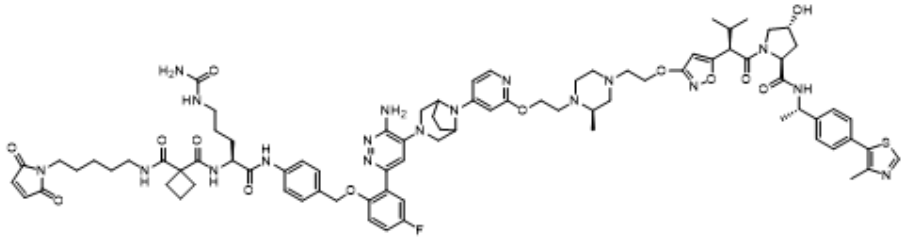
R_7 and R_8 are each independently hydrogen, halo, C_{1-5} alkyl, C_{1-5} alkoxy or hydroxy; and

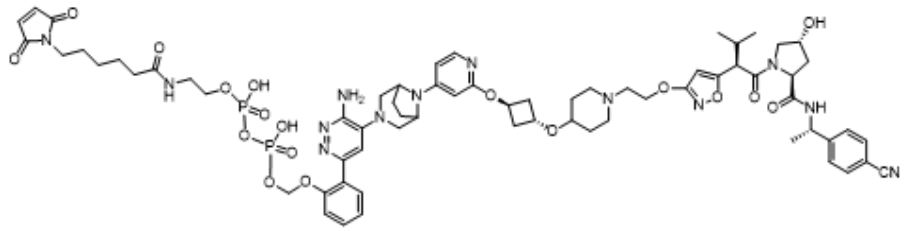
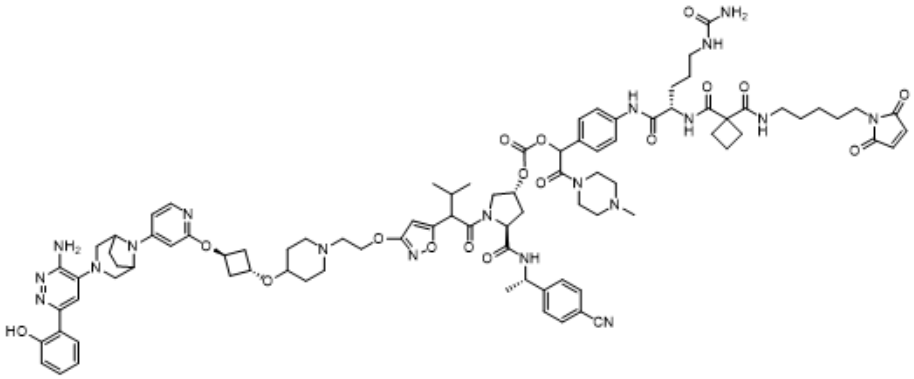
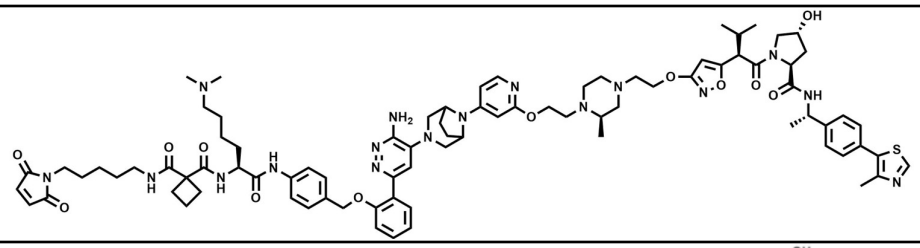
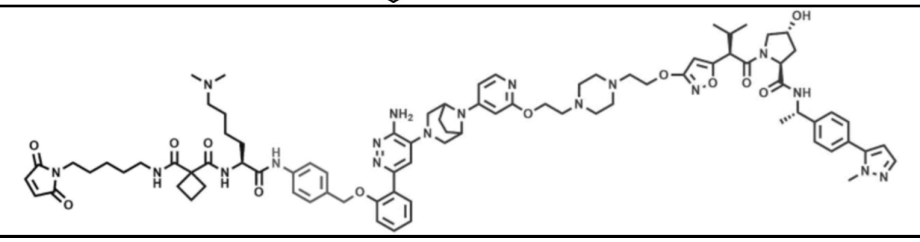
p has a value from about 1 to about 16.

In another aspect, the diclsoure provides a compound, selected from the group consisting of:

	L1-CIDE-BRM1-10

	L1-CIDE-BRM1-9
	L1-CIDE-BRM1-19
	L1-CIDE-BRM1-13
	L1-CIDE-BRM1-20
	L1-CIDE-BRM1-11

	L1-CIDE-BRM1-12
	L1-CIDE-BRM1-14
	L1-CIDE-BRM1-7
	L1-CIDE-BRM1-8
	L1-CIDE-BRM1-16

	L1-CIDE-BRM1-17
	L1-CIDE-BRM1-18
	L1-CIDE-BRM1-21
	L1-CIDE-BRM1-22

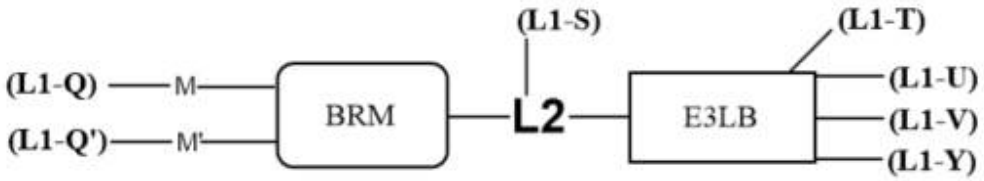
In another example, the subject matter described herein is directed to an Ab-CIDE having the chemical structure:

$$Ab\text{-}(L1\text{-}D)_p,$$

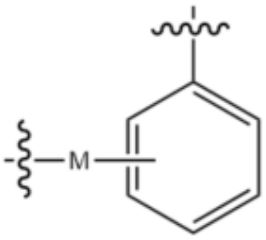
wherein,

Ab is an antibody;

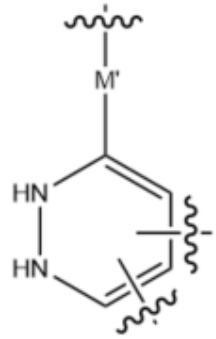
D is a CIDE, or prodrug thereof, having the structure:



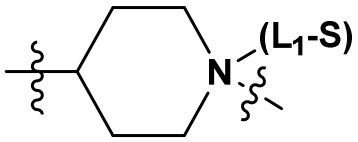
wherein L1 is attached at one attachment point selected from L1-Q, L1-Q', L1-S, L1-T, and optionally L1-U, L1-V and L1-Y, if present, wherein

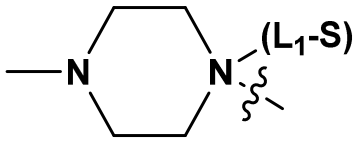
L1Q is at 

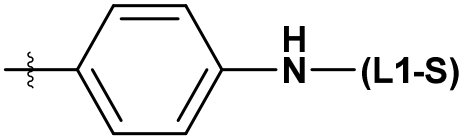
on BRM, wherein M is O;

L1-Q’ is at 

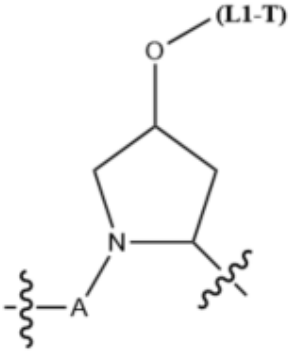
on BRM, wherein M’ is -NH;

L1-S is at 

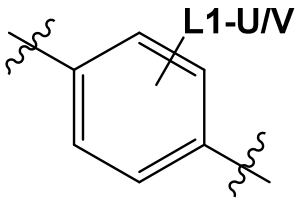
, 

or 

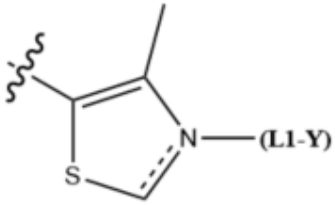
on L2;

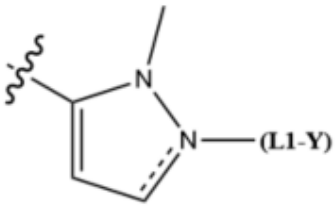


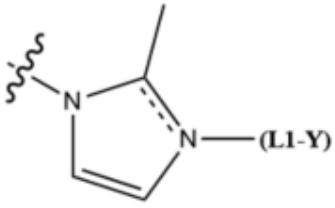
L1-T is at on E3LB, wherein, A is a group covalently bound to L2;



L1-U and L1-V are at on E3LB; and



L1-Y is at , 

or 

on E3LB, wherein, ---- is a single or double bond.

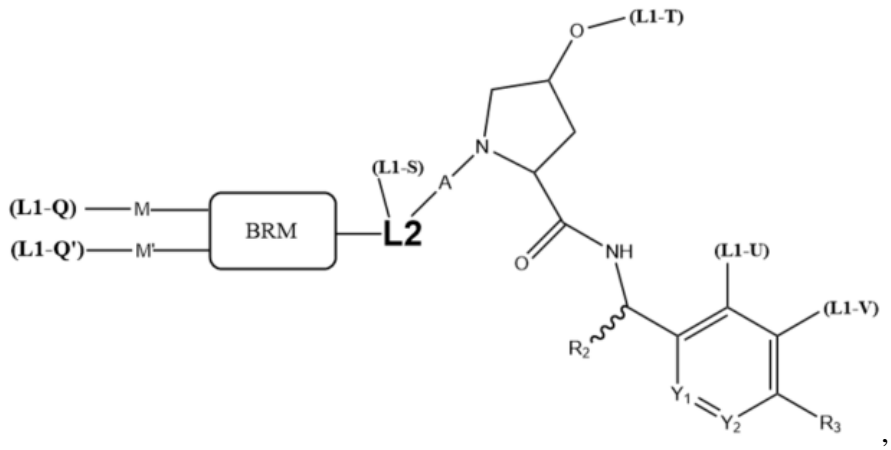
In another example, the subject matter described herein is directed to an Ab-CIDE having the chemical structure:

$Ab\text{-}(L1\text{-}D)_p$,

wherein,

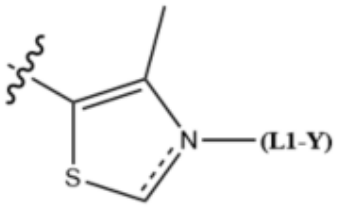
Ab is an antibody;

D is a CIDE, or prodrug thereof, having the structure:

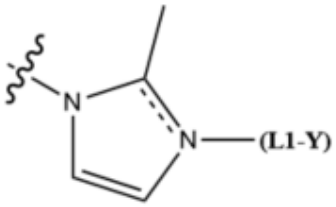


,

wherein:

R_3 is cyano, 

, or



,

wherein, ---- is a single or double bond.

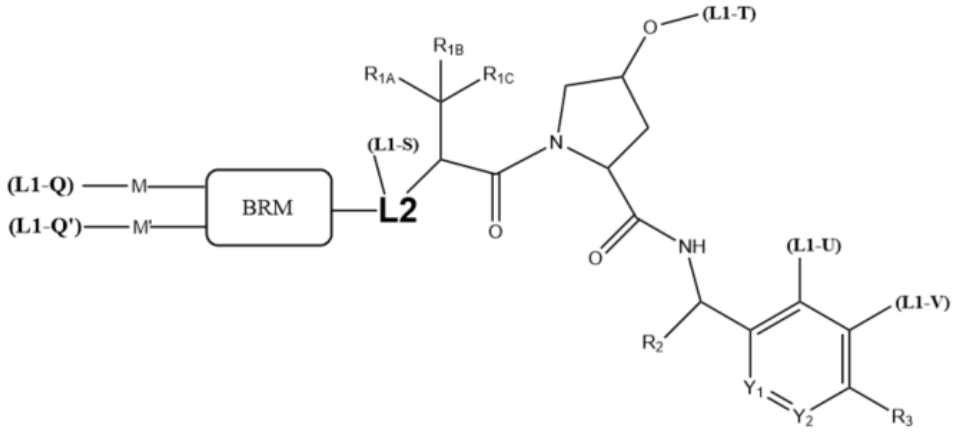
In another example, the subject matter described herein is directed to an Ab-CIDE having the chemical structure:

$$Ab\text{-}(L1\text{-}D)_p,$$

wherein,

Ab is an antibody;

D is a CIDE, or prodrug thereof, having the structure:



,

wherein, R_{1A}, R_{1B} and R_{1C} are each independently hydrogen, or $C_{1\text{-}5}$ alkyl; or two of R_{1A} , R_{1B} and R_{1C} together with the carbon to which each is attached form a $C_{1\text{-}5}$ cycloalkyl.

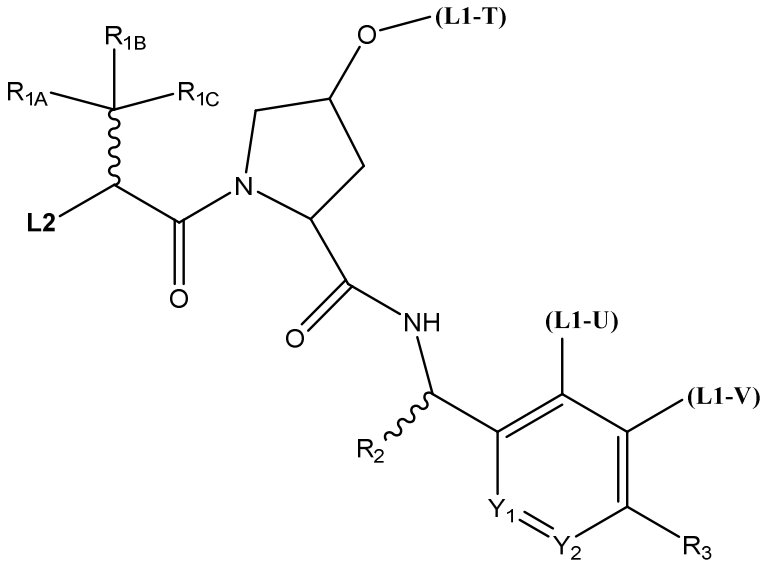
In another example, the subject matter described herein is directed to an Ab-CIDE having the chemical structure:

$$Ab—(L1—D)_p,$$

wherein,

D is a CIDE having the structure E3LB—L2—PB;

E3LB is covalently bound to L2, said E3LB having the formula:



E3LB

wherein,

R_{1A}, R_{1B} and R_{1C} are each independently hydrogen, or $C_{1\text{-}5}$ alkyl; or two of R_{1A} , R_{1B} and R_{1C} together with the carbon to which each is attached form a $C_{1\text{-}5}$ cycloalkyl;

p has a value from about 1 to about 8.

Another example of the subject matter described herein is a pharmaceutical composition comprising an Ab-CIDE, and one or more pharmaceutically acceptable excipients. In one aspect, the disclosure provides a pharmaceutical composition comprising the conjugate as disclosed herein, and one or more pharmaceutically acceptable excipients.

Another example of the subject matter described herein is the use of an Ab-CIDE in methods of treating conditions and diseases by administering to a subject a pharmaceutical composition comprising an Ab-CIDE. In one aspect, the disclosure provides use of the conjugate as disclosed herein or the pharmaceutical composition as disclosed herein in the preparation of a medicament for treating a disease in a subject in need thereof, wherein the disease is a cancer. In another aspect, the disclosure provides use of the conjugate as disclosed herein or the pharmaceutical composition as disclosed herein in the preparation of a medicament for reducing the level of BRM protein in a subject.

Another example of the subject matter described herein is a method of making an Ab-CIDE.

Another example of the subject matter described herein is an article of manufacture comprising a pharmaceutical composition comprising an Ab-CIDE, a container, and a package insert or label indicating that the pharmaceutical composition can be used to treat a disease or condition.

Yet other examples are also fully described herein.

BRIEF DESCRIPTION OF THE FIGURES

Figure 1A and 1B shows an exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-1 is active in cell-based assays.

Figure 2A and 2B shows an exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-3 is active in cell-based assays.

Figure 3A-3L shows dose and antigen-dependent anti-tumor activity of an exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-1.

Figure 4A-4L shows dose and antigen-dependent anti-tumor activity of an exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-3. The data are in relative contrast to those of Ab-CIDE Ab-L1a-CIDE-BRM1-1.

Figure 5 shows that BRM and BRG1 degradation correlate with anti-tumor activity of an exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-1.

Figure 6 shows that BRM and BRG1 degradation with anti-tumor activity of an exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-3 is less correlative. All lanes are for Ab-CIDE Ab-L1a-CIDE-BRM1-3 except ** indicates the lane for Ab-CIDE-L1a-BRM1-1.

Figure 7 shows that the antibody linking strategy can modulate the activity of the CIDE. The data show that the exemplary Ab-CIDE Ab-L1a-CIDE-BRM1-1 provides stronger BRM degradation than unconjugated CIDE-BRM1-3, although CIDE-BRM1-3 is

What is Claimed is:

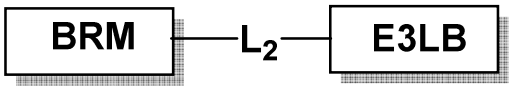
1. A conjugate having the structure:

$$Ab\text{-}(L1\text{-}D)_p,$$

wherein,

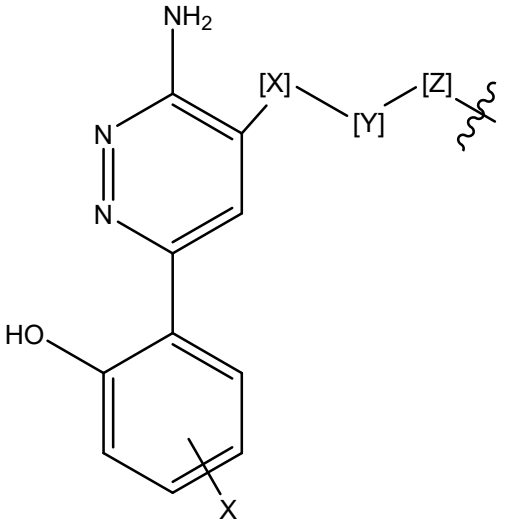
Ab is an antibody;

D is a CIDE, or prodrug thereof, the CIDE having the structure:

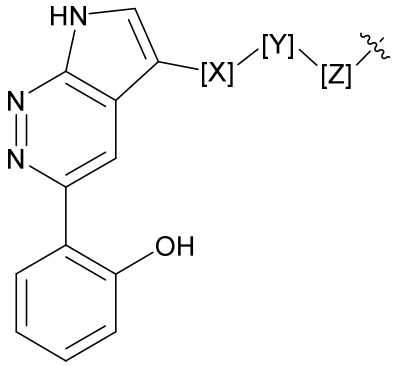


wherein,

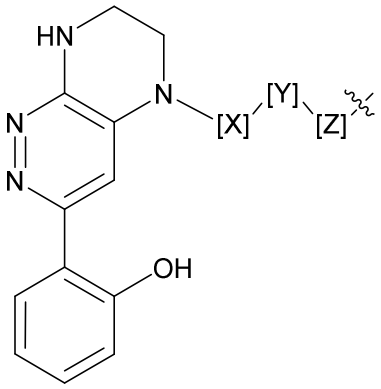
BRM is a residue of a BRM-binding compound, wherein the residue of a BRM-binding compound is a compound of formula (I-A), formula (I-B), formula (I-C), formula (I-D), or formula (I-E):



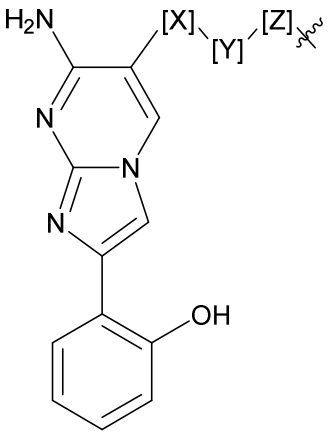
(I-A),



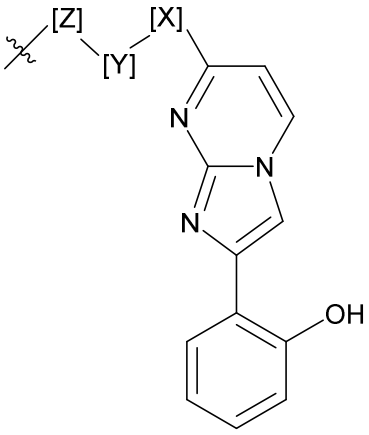
(I-B),



(I-C),



(I-D), wherein X is hydrogen or halogen; or



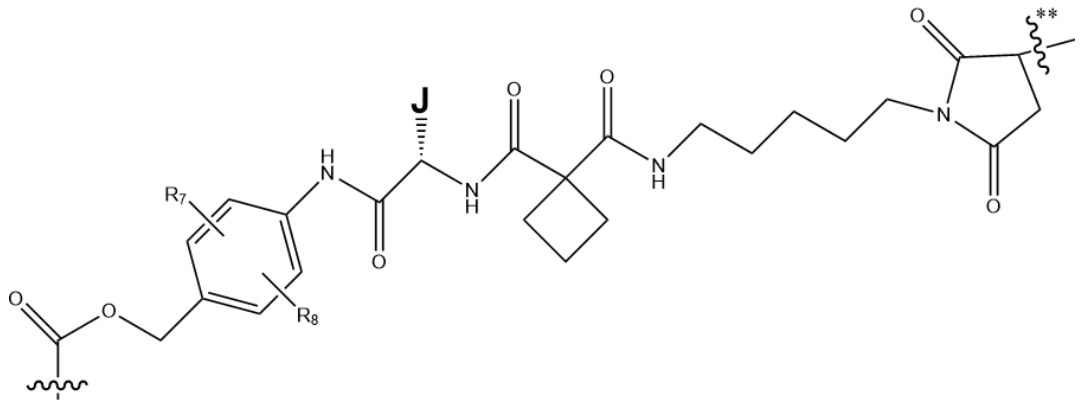
(I-E);

E3LB is a residue of an E3 ligase-binding compound, and

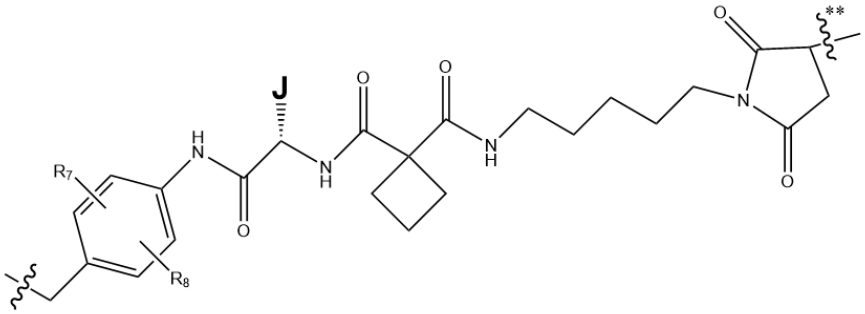
L2 is a linker covalently linking BRM and E3LB;

L1 is a linker covalently linking Ab to BRM, E3LB or L2;

wherein L1 is selected from the group consisting of:



, and



wherein,

J is —CH_2-CH_2-CH_2-NH-C(O)-NH_2; —CH_2-CH_2-CH_2-CH_2-NH_2; —CH_2-CH_2-CH_2-CH_2-NH-CH_3; or —CH_2-CH_2-CH_2-CH_2-N(CH_3)$_2$; and

R_7 and R_8 are each independently hydrogen, halo, C_{1-5} alkyl, C_{1-5} alkoxy or hydroxy; and

p has a value from about 1 to about 16.

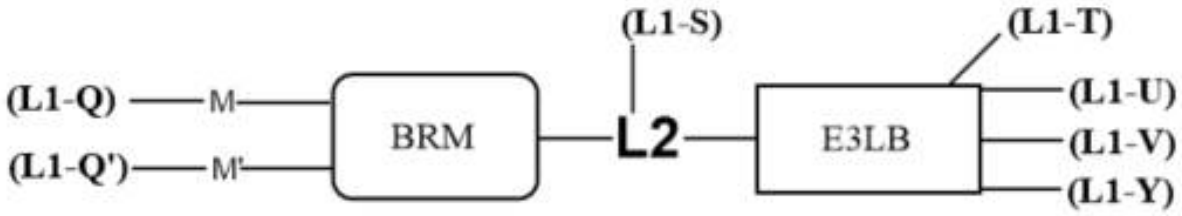
2. The conjugate of claim 1, wherein L1 is covalently bound to E3LB.

3. The conjugate of claim 2, wherein the prodrug is a CIDE having a phosphate moiety covalently bound to BRM.

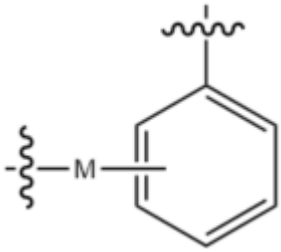
4. The conjugate of claim 1, wherein L1 is covalently bound to BRM.

5. The conjugate of claim 4, wherein the prodrug is a CIDE having a phosphate moiety is covalently bound to E3LB.

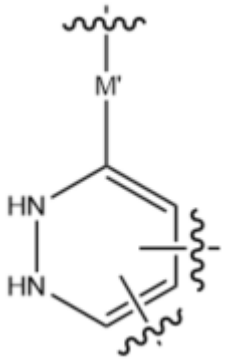
6. The conjugate of claim 1, wherein D has the structure



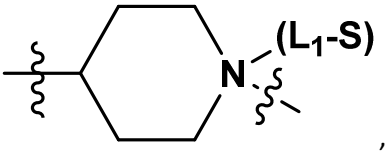
wherein L1 is attached at one attachment point selected from L1-Q, L1-Q', L1-S, L1-T, and optionally L1-U, L1-V and L1-Y, if present, wherein

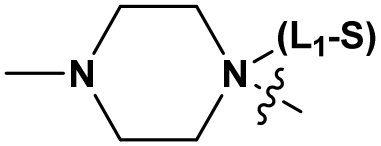


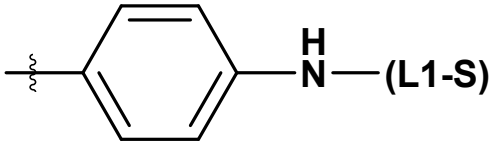
L1Q is at on BRM, wherein M is O;



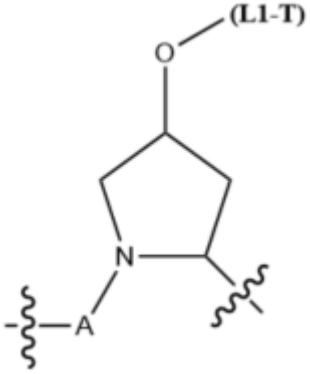
L1-Q' is at on BRM, wherein M' is -NH;

L1-S is at 

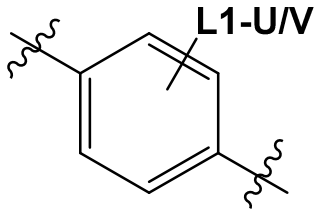
, 

or 

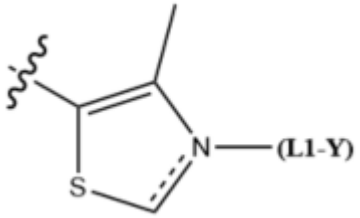
on L2;

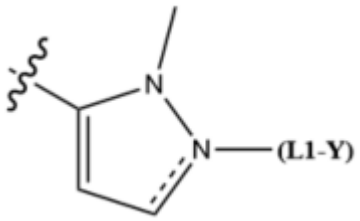
L1-T is at 

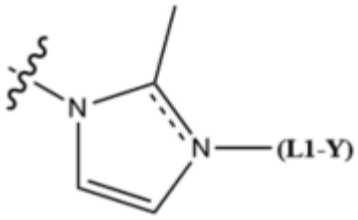
on E3LB, wherein, A is a group covalently bound to L2;

L1-U and L1-V are at 

on E3LB; and

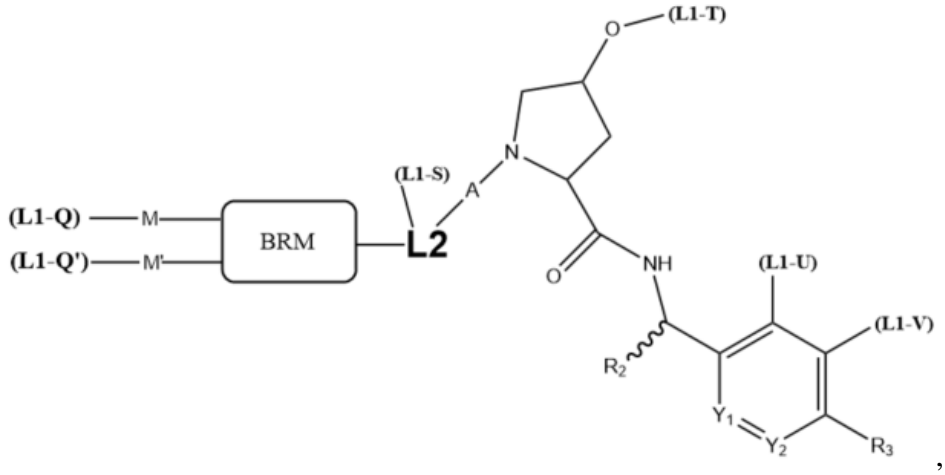
L1-Y is at 

, 

or 

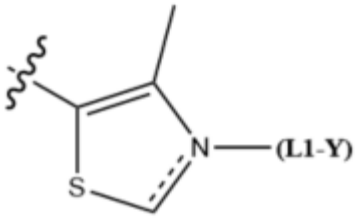
on E3LB, wherein, ---- is a single or double bond.

7. The conjugate of claim 6, wherein D has the structure:

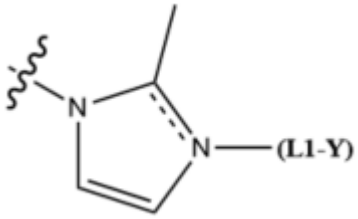


,

wherein:

R_3 is cyano, 

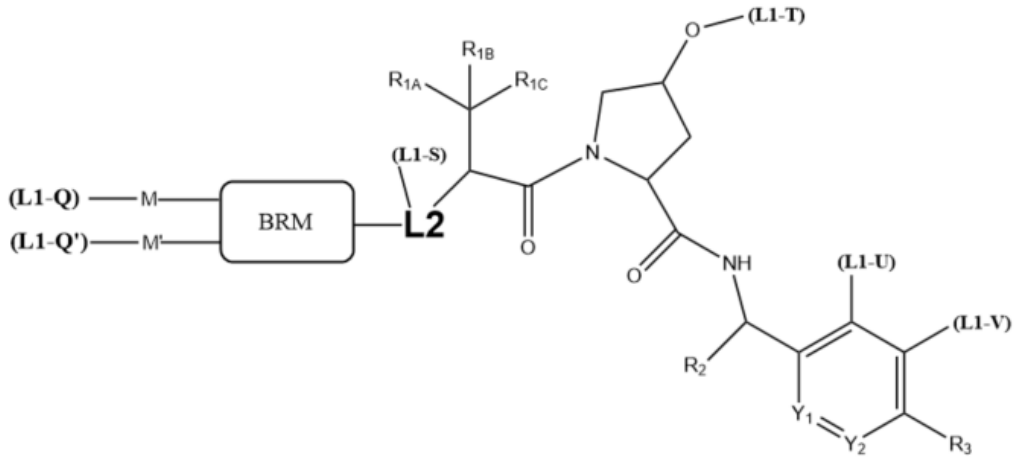
, or



,

wherein, ---- is a single or double bond.

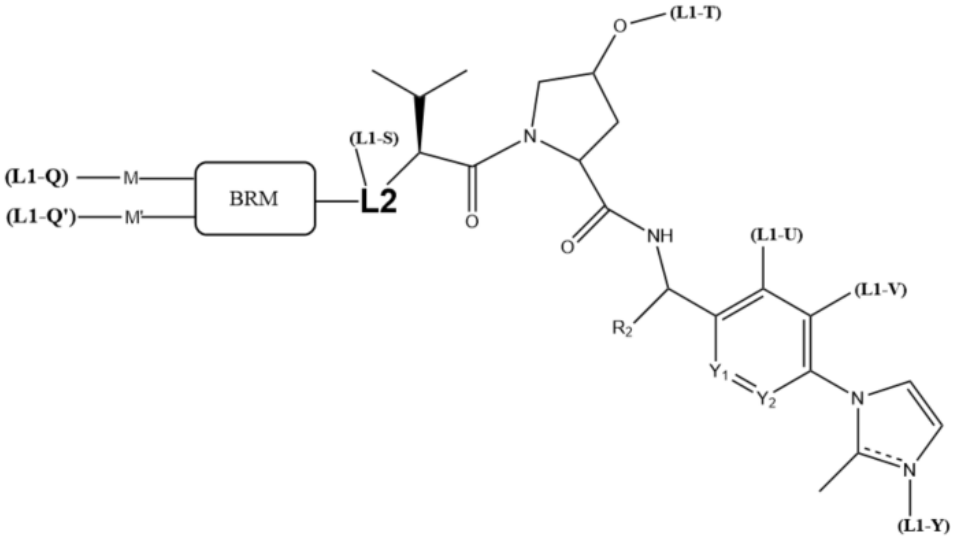
8. The conjugate of claim 6, wherein D has the structure:



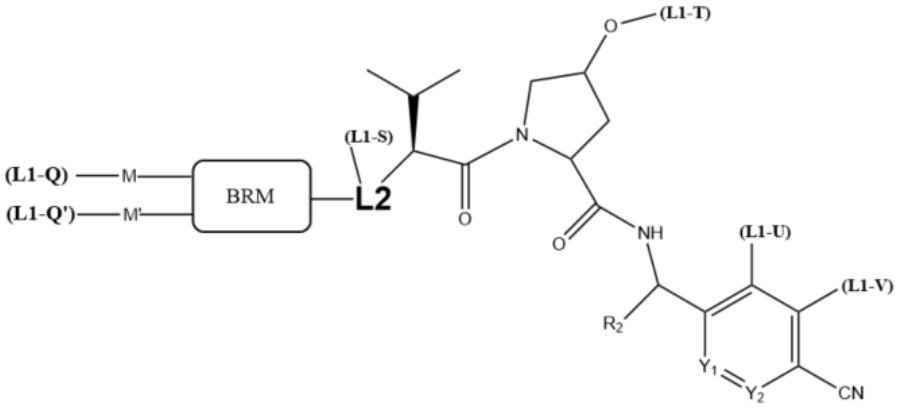
,

wherein, R_{1A}, R_{1B} and R_{1C} are each independently hydrogen or methyl.

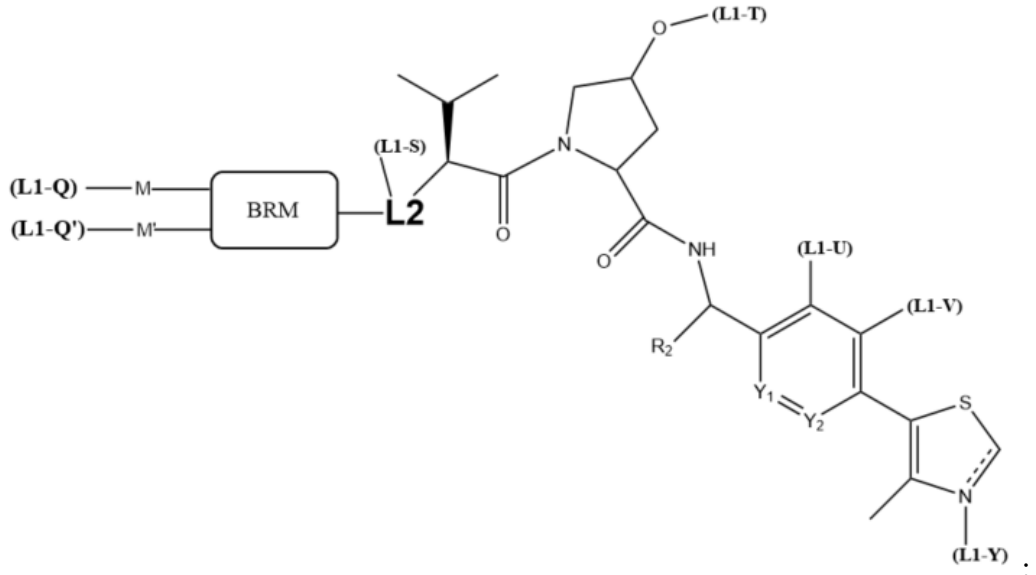
9. The conjugate of claim 6, wherein D has the structure:



DAC4
,



DAC5
, or

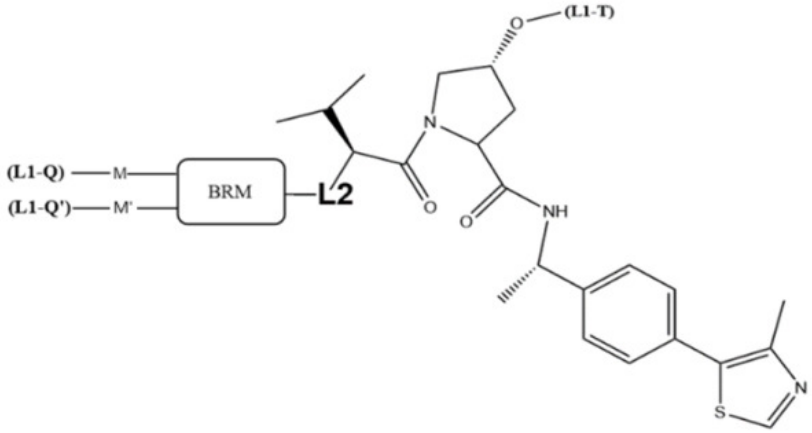


DAC6

;

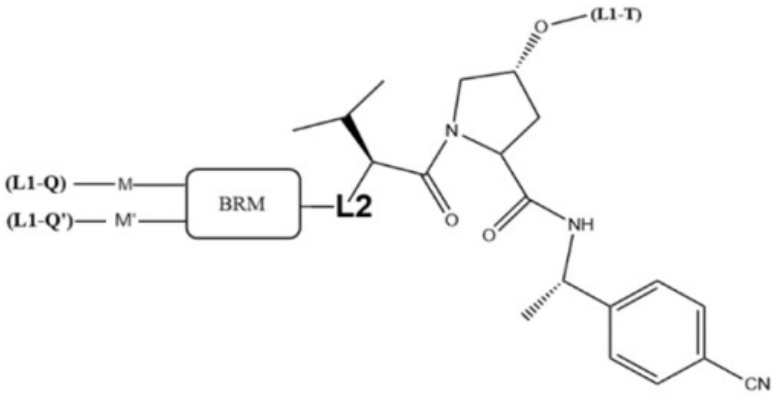
and wherein R_2 is methyl and Y_1 and Y_2 are each -CH.

10. The conjugate of claim 6, wherein D has the structure:



DAC7

, or

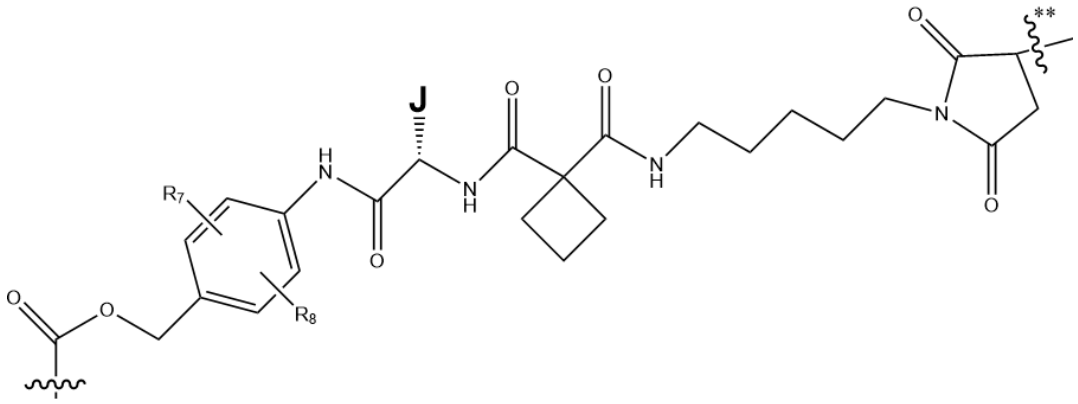


DAC8

.

11. The conjugate of claim 1, wherein J is —CH_2-CH_2-CH_2-NH-C(O)-NH_2 or —CH_2-CH_2-CH_2-CH_2-N$(CH_3)_2$.

12. The conjugate of claim 1, wherein L1 has the structure:



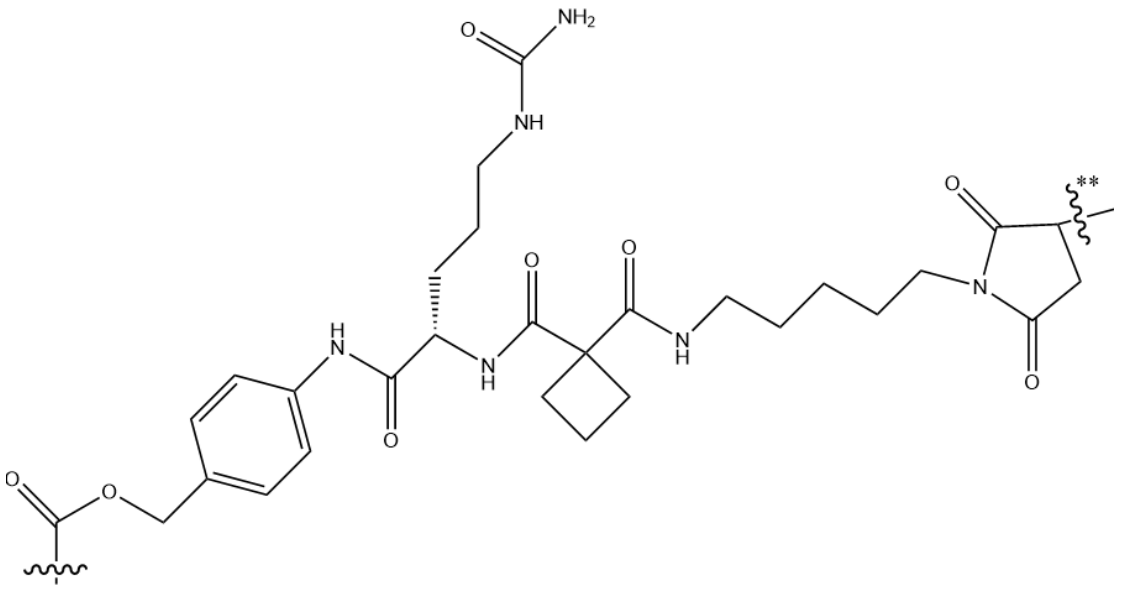
,

wherein,

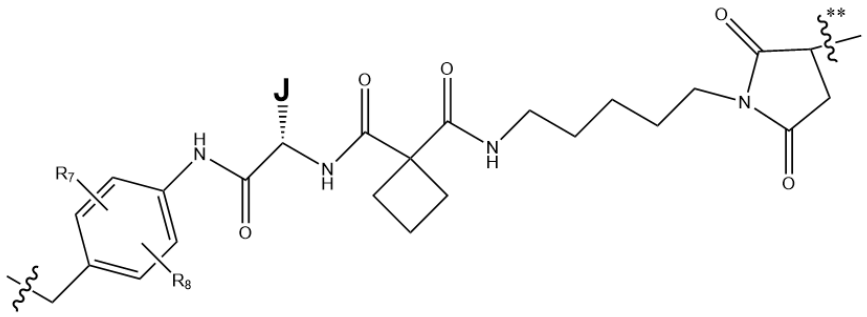
J is —CH_2-CH_2-CH_2-NH-C(O)-NH_2; —CH_2-CH_2-CH_2-CH_2-NH_2; —CH_2-CH_2-CH_2-CH_2-NH-CH_3; or —CH_2-CH_2-CH_2-CH_2-N$(CH_3)_2$; and

R_7 and R_8 are each independently hydrogen, halo, C_{1-5} alkyl, C_{1-5} alkoxy or hydroxy.

13. The conjugate of claim 12, wherein L1 has the structure:



14. The conjugate of claim 1, wherein L1 has the structure:

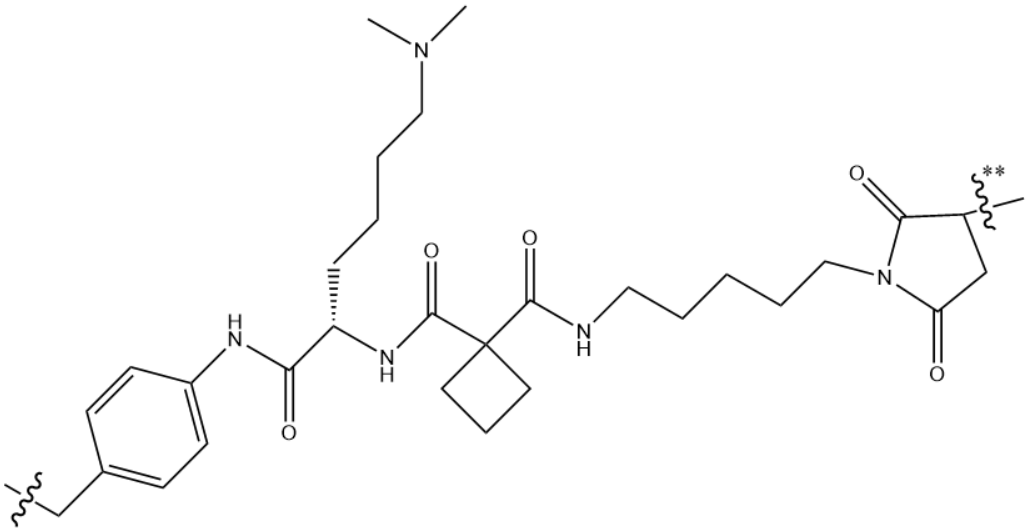


wherein,

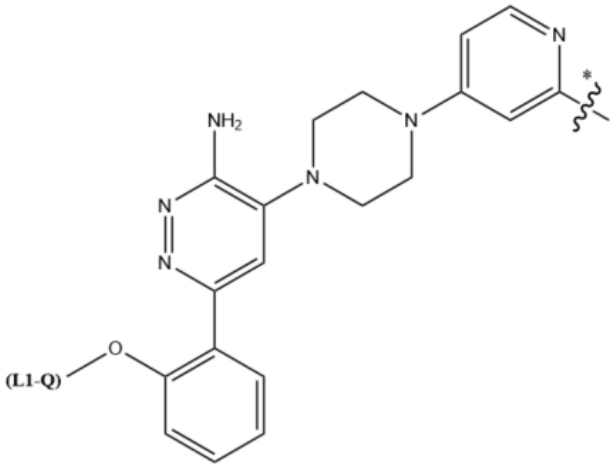
J is —CH_2-CH_2-CH_2-NH-C(O)-NH_2; —CH_2-CH_2-CH_2-CH_2-NH_2; —CH_2-CH_2-CH_2-CH_2-NH-CH_3; or —CH_2-CH_2-CH_2-CH_2-$N(CH_3)_2$; and

R_7 and R_8 are each independently hydrogen, halo, C_{1-5} alkyl, C_{1-5} alkoxy or hydroxy.

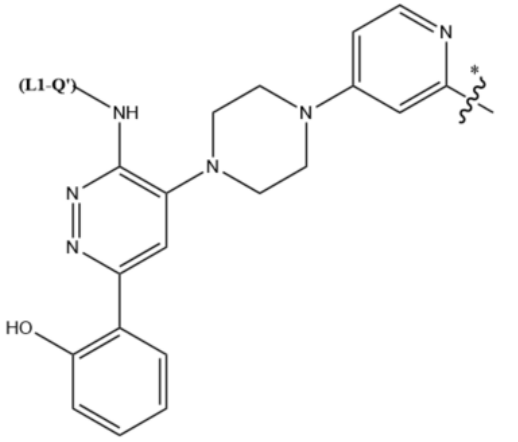
15. The conjugate of claim 14, wherein L1 has the structure:



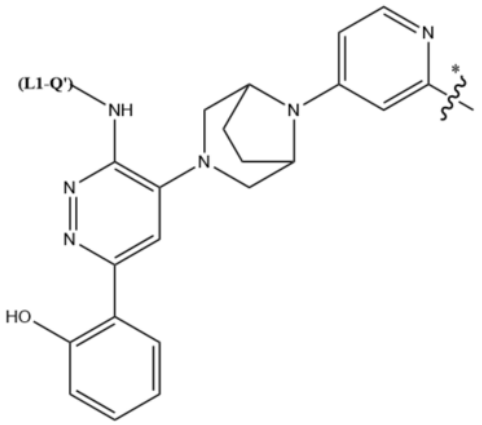
16. The conjugate of claim 1, wherein BRM is a residue of:



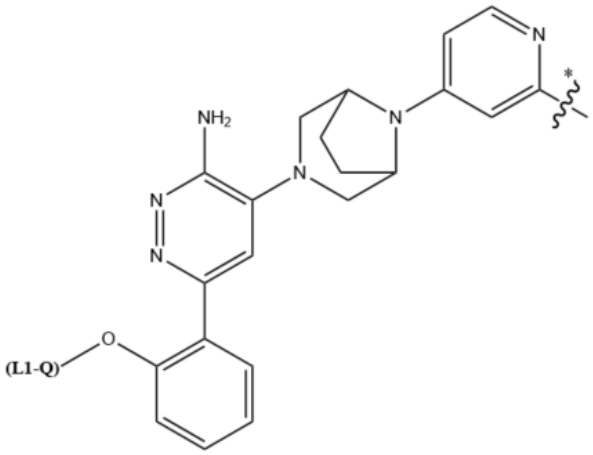
,



,



, and

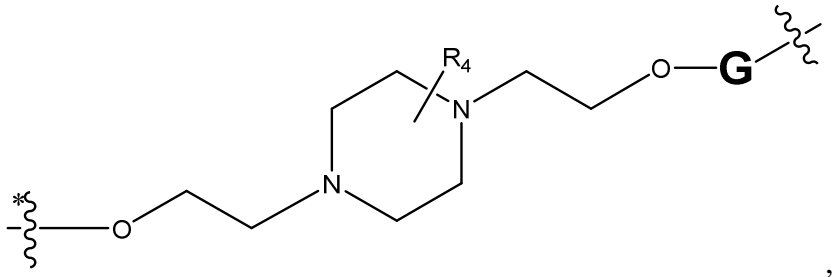


,

wherein  is the point of attachment to L2.

17. The conjugate of claim 1, wherein:

L2 has the formula of structure L2a, L2b, or L2c:

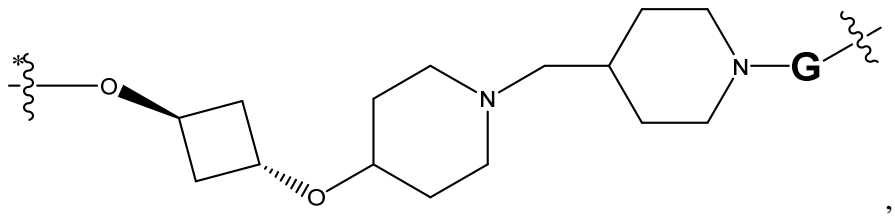


L2a

,

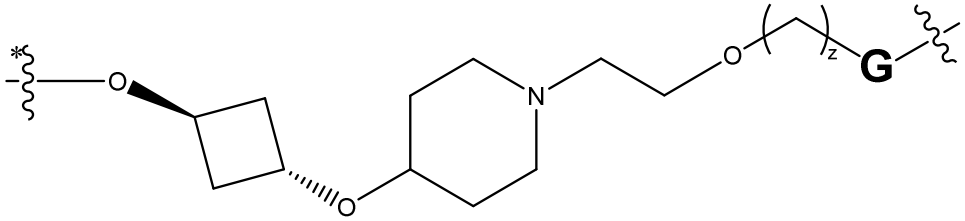
wherein,

R_4 is hydrogen or methyl,



L2b

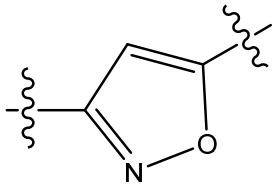
, or



L2c

wherein,

z is one or zero,

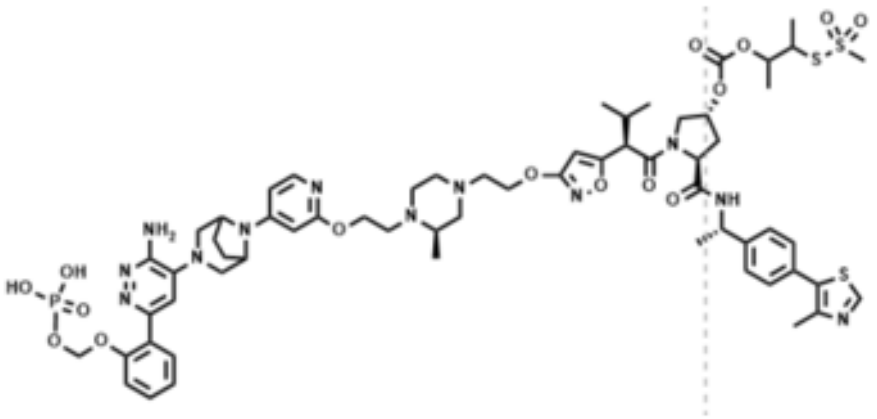
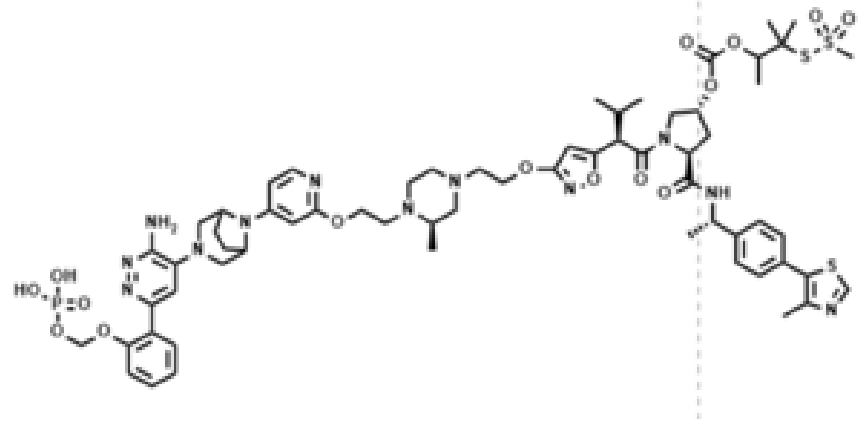
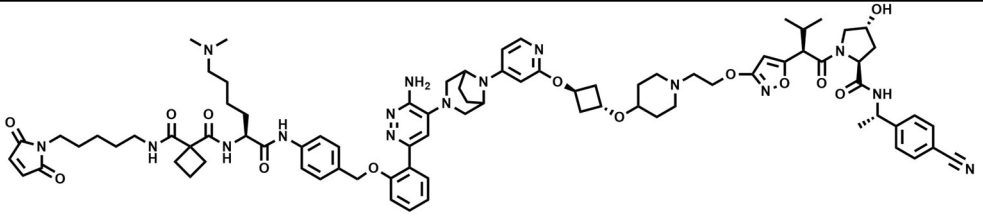
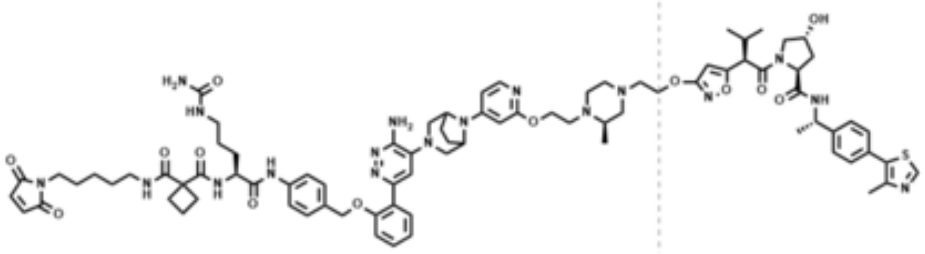
G is  or —C(O)NH—; and,

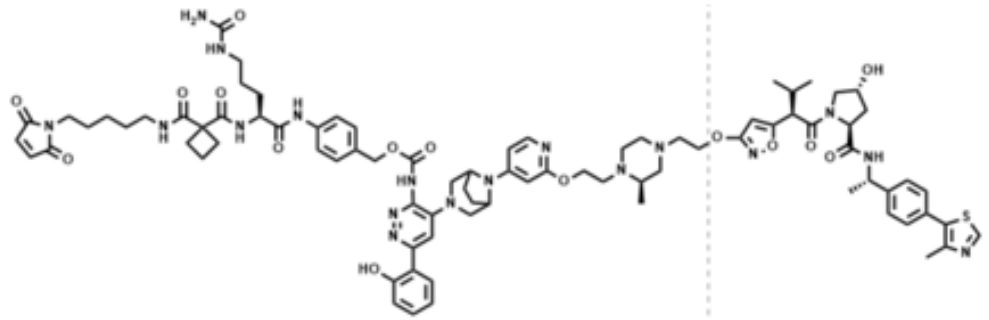
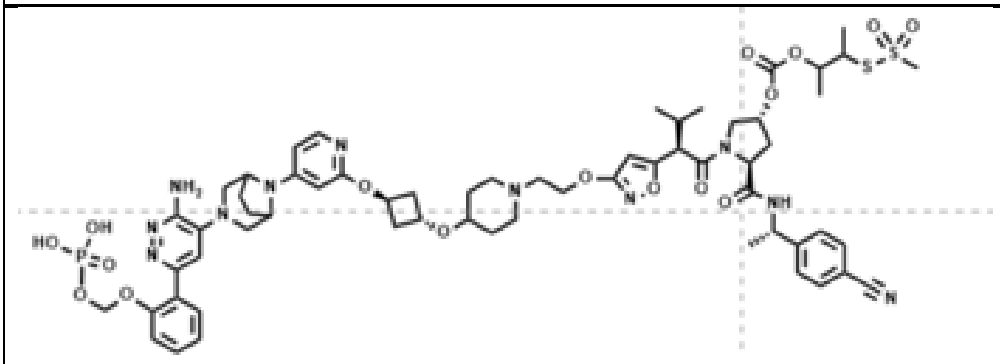
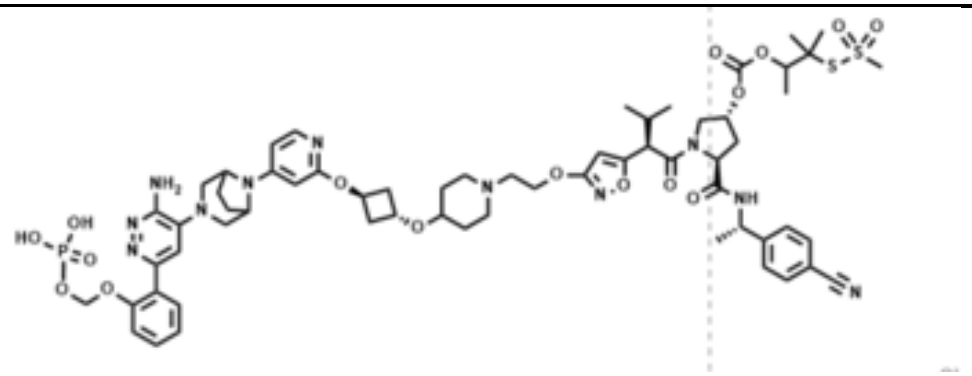
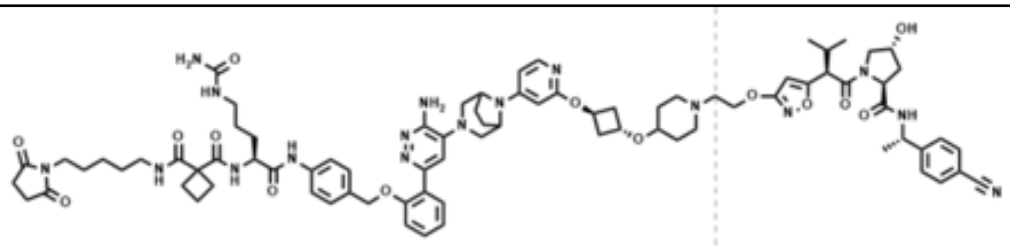
 is the point of attachment to BRM.

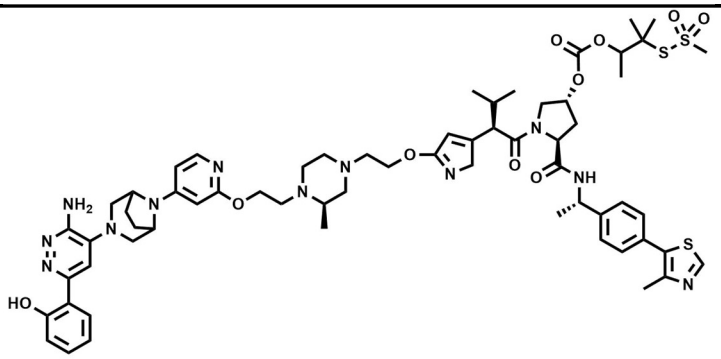
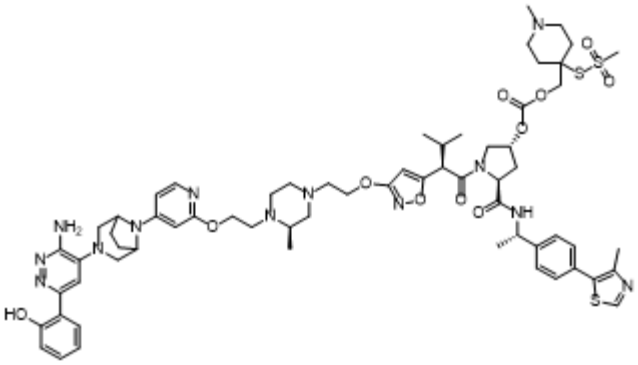
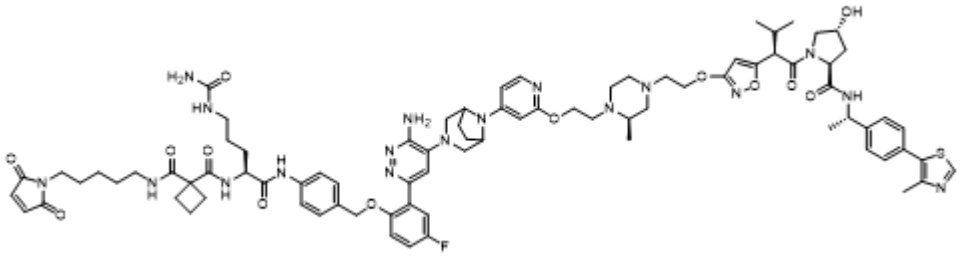
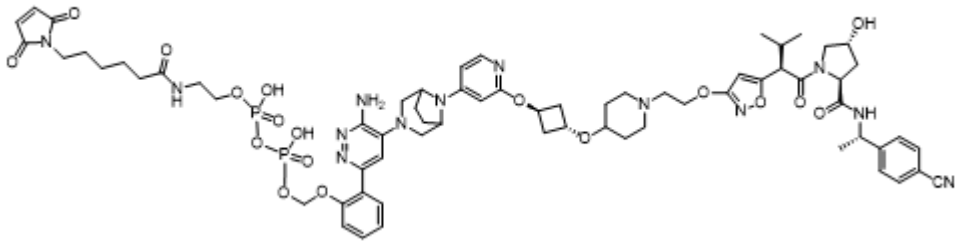
18. The conjugate of claim 17, wherein R_4 is hydrogen.

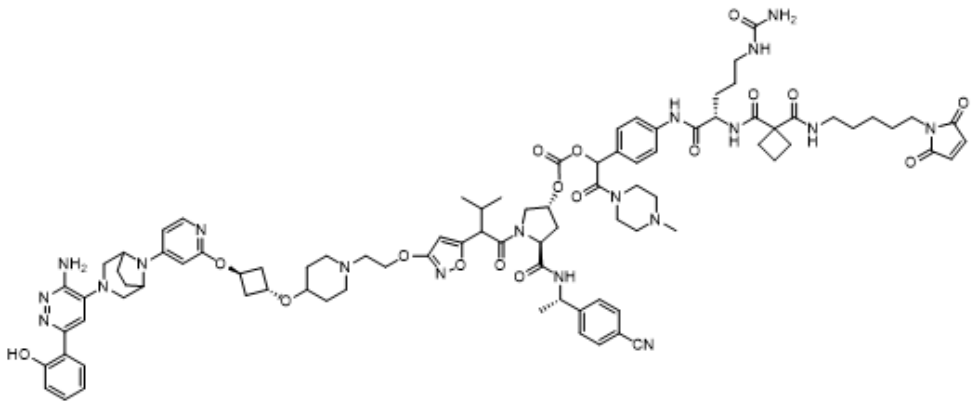
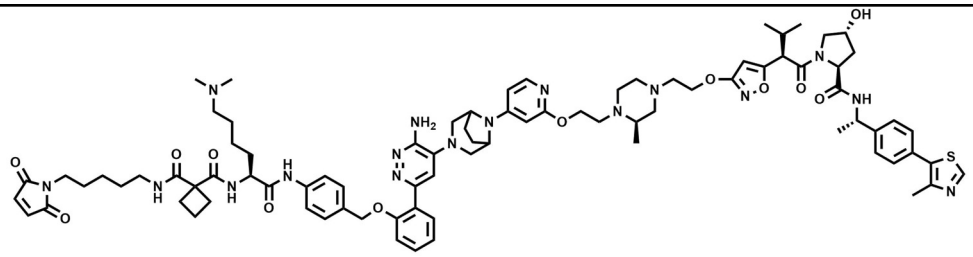
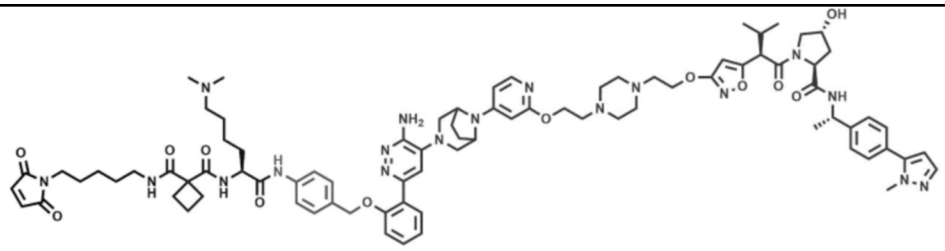
19. The conjugate of claim 17, wherein R_4 is methyl.

20. A compound, selected from the group consisting of:

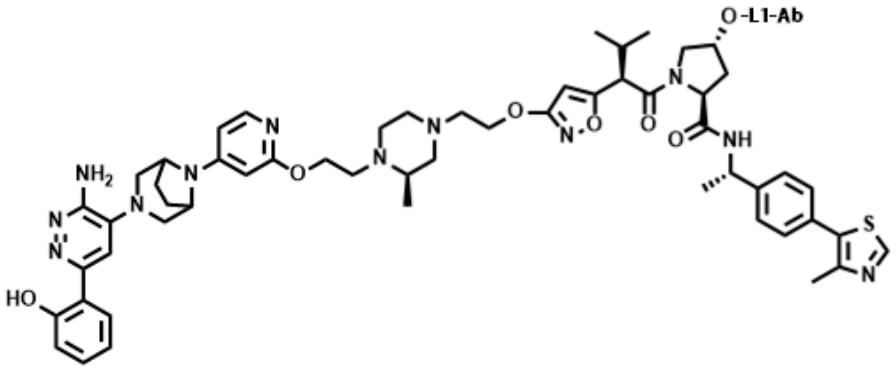
	L1-CIDE-BRM1-10
	L1-CIDE-BRM1-9
	L1-CIDE-BRM1-19
	L1-CIDE-BRM1-13

	L1-CIDE-BRM1-20
	L1-CIDE-BRM1-11
	L1-CIDE-BRM1-12
	L1-CIDE-BRM1-14

	L1-CIDE-BRM1-7
	L1-CIDE-BRM1-8
	L1-CIDE-BRM1-16
	L1-CIDE-BRM1-17

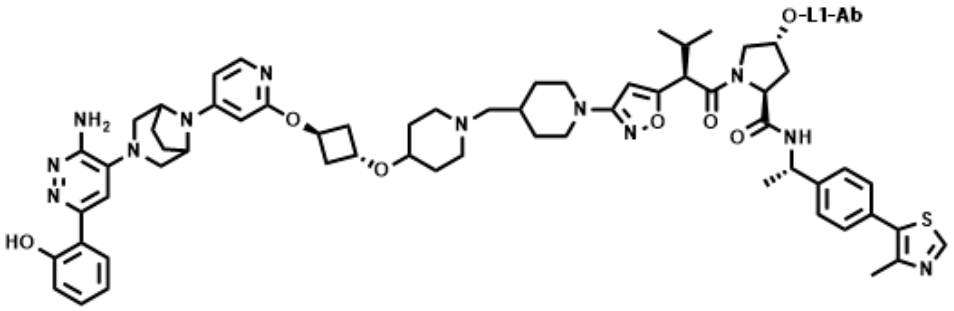
	L1-CIDE-BRM1-18
	L1-CIDE-BRM1-21
	L1-CIDE-BRM1-22

21. The conjugate of claim 1, selected from the group consisting of:

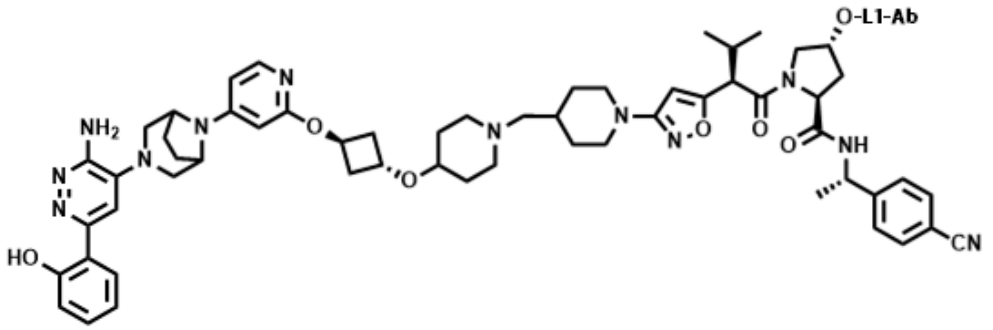


1-L2a

,

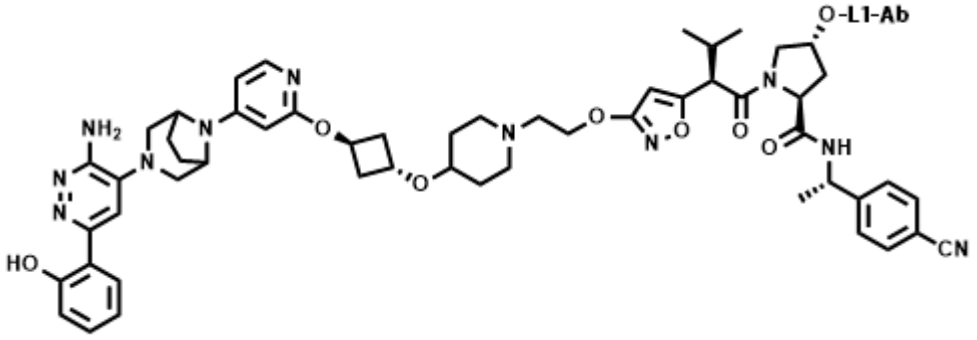


2-L2b,



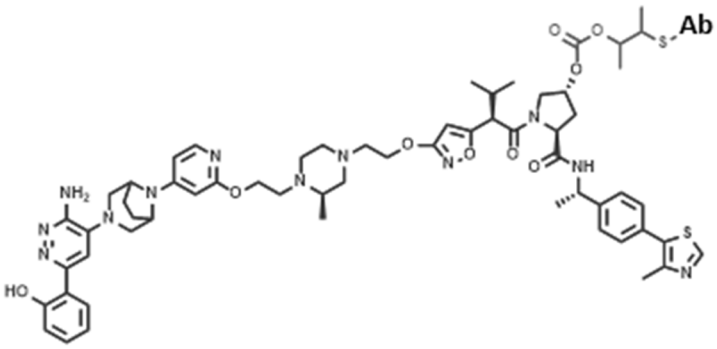
3-L2b,

and

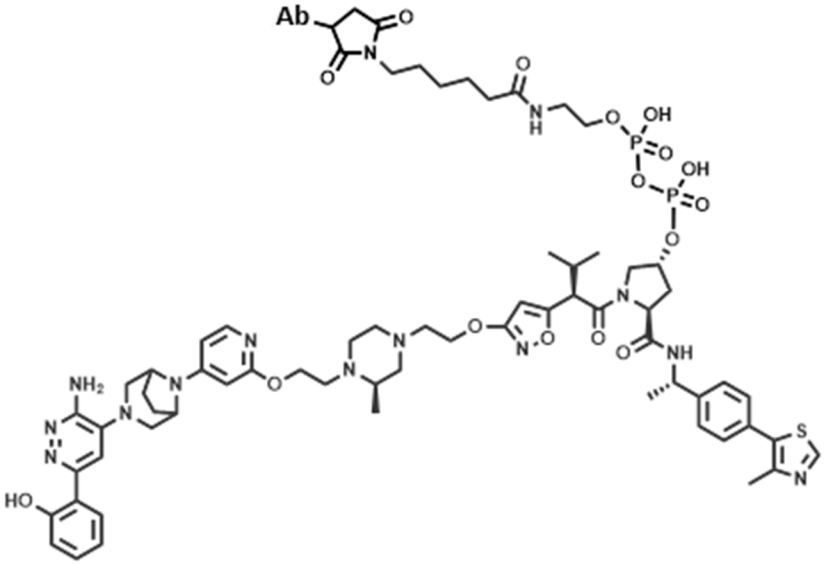


4-L2c.

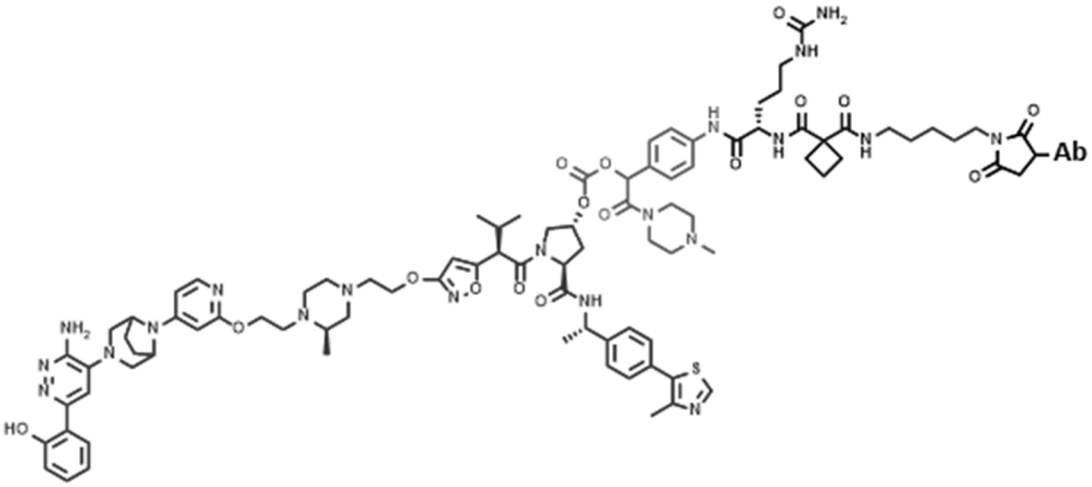
22. The conjugate of claim 1, selected from the group consisting of:



,



, and



.

23. The conjugate of claim 1, wherein p has a value from about 5 to about 14.

24. The conjugate of claim 1, wherein p has a value from about 5 to about 10.

25. A pharmaceutical composition comprising the conjugate of any one of claims 1-19 and 21-24, and one or more pharmaceutically acceptable excipients.

26. Use of the conjugate according to any one of claims 1-19 and 21-24 or the pharmaceutical composition of claim 25 in the preparation of a medicament for treating a disease in a subject in need thereof, wherein the disease is a cancer.

27. Use of the conjugate according to any one of claims 1-19 and 21-24 or the pharmaceutical composition of claim 25 in the preparation of a medicament for reducing the level of BRM protein in a subject.