

WHAT IS CLAIMED IS:

1. A method for determining whether a subject is or will become less responsive to therapy with a first- or second-generation androgen receptor (AR) antagonist, comprising:
 - (a) testing a sample containing a nucleic acid molecule encoding an AR polypeptide from the subject to determine whether the encoded AR polypeptide is modified at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1; and
 - (b) characterizing the subject as resistant or will become resistant to therapy with a first- or second-generation AR antagonist if the subject has the modification.
2. The method of claim 1, wherein the subject has been administered a first- or second-generation AR antagonist for the treatment of a cancer.
3. The method of claim 2, wherein the cancer is prostate cancer.
4. A method for optimizing the therapy of a subject receiving a first- or second-generation AR antagonist for treatment of a cancer, comprising:
 - (a) testing a sample containing a nucleic acid molecule encoding an AR polypeptide from the subject to determine whether the encoded AR polypeptide is modified at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1; and
 - (b) discontinuing treatment with the first- or second-generation AR antagonist if the subject has the modification or continuing treatment with a first- or second-generation AR antagonist if the subject does not have the modification.
5. A method for selecting a subject for therapy with a third-generation AR antagonist, comprising:
 - (a) testing a sample containing a nucleic acid molecule encoding an AR polypeptide from the subject to determine whether the encoded AR polypeptide is modified at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1; and
 - (b) characterizing the subject as a candidate for therapy with a third-generation AR antagonist if the subject has the modification.
6. The method of claim 2 or 3, further comprising discontinuing treatment with a first- or second-generation AR antagonist if the subject has the modification or continuing treatment with a first- or second-generation AR antagonist if the subject does not have the modification.

7. The method of any of claims 1-6, further comprising administering third-generation AR antagonist that inhibits the modified receptor if the subject has the modification.

8. The method of any of claims 1-7, wherein the modification comprises a substitution or a deletion of the amino acid at amino acid position 876 in the AR polypeptide.

9. The method of claim 8, wherein the modification is a substitution of phenylalanine to an amino acid selected from among leucine, isoleucine, valine, alanine, glycine, methionine, serine, threonine, cysteine, tryptophan, lysine, arginine, histidine, proline, tyrosine, asparagine, glutamine, aspartic acid and glutamic acid at amino acid position 876 of the AR polypeptide.

10. The method of claim 8, wherein the modification is a substitution of phenylalanine to an amino acid selected from among glycine, alanine, valine, leucine, and isoleucine at amino acid position 876 of the AR polypeptide.

11. The method of claim 9, wherein the modification is a substitution of phenylalanine to leucine at amino acid position 876 of the AR polypeptide.

12. The method of any of claims 1-8, wherein the modification comprises a deletion of nucleic acid encoding amino acid position 876 of the AR polypeptide.

13. The method of any of claims 1-12, wherein the nucleic acid encoding the modified AR polypeptide has (i) a mutation of thymine (t) to cytosine (c) at a nucleic acid position corresponding to nucleic acid position 2626 in the sequence of nucleotides set forth in SEQ ID NO: 18; (ii) a mutation of cytosine (c) to adenine (a) at nucleic acid position corresponding to nucleic acid position 2628 in the sequence of nucleotides set forth in SEQ ID NO: 18; or (iii) a mutation of cytosine (c) to guanine (g) at nucleic acid position corresponding to nucleic acid position 2628 in the sequence of nucleotides set forth in SEQ ID NO: 18.

14. The method of claim 13, wherein the nucleic acid encodes leucine at amino acid position 876 of the AR polypeptide.

15. The method of any of claims 1-14, wherein the nucleic acid molecule is RNA or DNA.

16. The method of claim 14, wherein the DNA is genomic DNA.

17. The method of any of claims 1-14, wherein the method further comprises isolating mRNA from the RNA sample.

18. The method of any of claims 1-17, wherein testing comprises amplifying nucleic acid encoding amino acid position 876 of the AR polypeptide.

19. The method of claim 18, wherein amplification is by polymerase chain reaction (PCR).

20. The method of claim 19, wherein PCR amplification comprises using a pair of oligonucleotide primers that flank the region encoding amino acid position 876 of the AR polypeptide.

21. The method of any of claims 1-20, wherein the method comprises sequencing the amplified nucleic acid.

22. The method of any of claims 1-21, wherein testing comprises contacting the nucleic acid with a sequence specific nucleic acid probe, wherein the sequence specific nucleic acid probe:

(a) binds to nucleic acid encoding a modified receptor that is modified at amino acid position 876; and

(b) does not bind to nucleic acid encoding the wild-type receptor having phenylalanine at amino acid position 876.

23. The method of any of claims 1-22, wherein the sample is from a tumor cell sample from the subject.

24. The method of any of claims 1-22, wherein sample is from a tumor biopsy sample, a blood sample, a serum sample, a lymph sample, or a bone marrow aspirate.

25. The method of claim 23 or 24, wherein the sample contains circulating tumor cells (CTC) or disseminated tumor cells (DTC).

26. The method of any of claims 1-25, wherein the first- or second-generation AR antagonist inhibits a wild-type AR polypeptide by competitive antagonism.

27. The method of any of claims 1-26, wherein the second-generation AR antagonist is selected from among ARN-509, enzalutamide (MDV3100), and RD162.

28. The method of any of claims 1-27, wherein the subject has a disease or disorder selected from among cancer, an inflammatory disorder or a proliferative disorder.

29. The method of claim 28, wherein the subject has cancer.

30. The method of claim 29, wherein the cancer is a prostate cancer, a breast cancer, a bladder cancer, or a hepatocellular cancer.

31. The method of claim 30, wherein the cancer is prostate cancer.

32. The method of claim 31, wherein the subject has a castration resistant prostate cancer.

33. The method of any of claims 1-32, wherein the subject has a solid tumor.

34. The method of any of claims 1-33, wherein the subject is treated with the first- or second-generation AR antagonist prior to obtaining the sample.

35. The method of claim 34, wherein the sample is a sample obtained at 1 week, 2 weeks, 3 weeks, 1 month, 2 months, 3 months, 4 months, 5 months, 6 months, 7 months, 8 months, 9 months, 10 months, 11 months, 12 months, 14 months, 16 months, 18 months, 20 months, 22 months, or 24 months following the first administration of with the first- or second-generation AR antagonist.

36. The method of claim 34 or 35, wherein the sample is obtained 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 times over the course of treatment with the first- or second-generation AR antagonist.

37. The method of any of claims 1-36, wherein the subject is responsive the treatment with the first- or second-generation AR antagonist when it is first administered.

38. A method for screening compounds that antagonize a modified AR, comprising:

(a) expressing a modified AR in a cell, wherein the modified AR is modified at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1;

(b) contacting the cell with a test compound; and

(c) detecting the level of AR activity in the cell, wherein a decrease in activity indicates that the compound antagonizes the modified AR receptor.

39. The method of claim 38, wherein the test compound exhibits full antagonist activity toward the modified AR receptor.

40. The method of claim 38 or 39, wherein the test compound does not exhibit agonist activity toward the modified AR receptor.

41. The method of any of claims 38-40, wherein the modification is a substitution or deletion of the amino acid at position 876 of the AR polypeptide.

42. The method of claim 39, wherein the modification is a substitution of phenylalanine to an amino acid selected from among leucine, isoleucine, valine, alanine, glycine, methionine, serine, threonine, cysteine, tryptophan, lysine, arginine, histidine, proline, tyrosine, asparagine, glutamine, aspartic acid and glutamic acid at amino acid position 876 of the AR polypeptide.

43. The method of claim 42, wherein the modification is a substitution of phenylalanine to an amino acid selected from among glycine, alanine, valine, leucine, and isoleucine at amino acid position 876 of the AR polypeptide.

44. The method of claim 43, wherein the modification is a substitution of phenylalanine to leucine at amino acid position 876 of the AR polypeptide.

45. The method of any of claims 38-44, wherein the cell is deficient for the expression of wild-type AR, expresses a low level of wild-type AR, or expresses a modified AR receptor.

46. The method of any of claims 38-45, wherein the cell is selected from among HeLa, CV1, COS7, HepG2, HEK-293, DU145, PC3, TSY-PR1, LNCaP, CWR, VCaP and LAPC4.

47. The method of any of claims 38-46, wherein the cell comprises a reporter gene operably linked to an androgen responsive promoter.

48. The method of claim 47, wherein activity is determined by analyzing the expression of the reporter gene.

49. The method of claim 47 or claim 48, wherein the promoter comprises androgen response element.

50. The method of claim 49, wherein the androgen response element is 4xARE or a probasin element.

51. The method of any of claims 38-50, wherein the promoter is a probasin, a prostate specific antigen, MMTV LTR, FASN, STEAP4, TMPRSS2, ORM1, or NKX3.1 promoter.

52. The method of any of claims 38-51, wherein the reporter gene encodes a protein selected from among a luciferase, a fluorescent protein, a bioluminescent protein, or an enzyme.

53. An isolated AR polypeptide or a variant thereof having AR activity comprising a modification at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1, wherein the modification confers resistance to an AR antagonist on the modified AR polypeptide or variant.

54. The isolated AR polypeptide of claim 53, wherein the modification comprises substitution of the amino acid at position 876 compared to a wild-type AR set forth in SEQ ID NO: 1.

55. The isolated AR polypeptide of claim 53 or claim 54, wherein the substitution is F876L.

56. The isolated AR polypeptide of claim 55 having the sequence of amino acids set forth in SEQ ID NO: 5.

57. The isolated AR polypeptide of claim 53, wherein the modification comprises a deletion of amino acid position 876.

58. The isolated AR polypeptide of any of claims 53-57, wherein the polypeptide comprises a substitution of the amino acid at position 876 compared to a wild-type AR set forth in SEQ ID NO: 1 and one or more additional amino acid substitutions.

59. The isolated AR polypeptide of claim 58, wherein substitution at amino acid at position 876 is F876L.

60. The isolated AR polypeptide of any of claims 58-59, wherein the one or more additional amino acid substitutions is selected from among one or more amino acid substitutions associated with castration resistant prostate cancer.

61. The isolated AR polypeptide of any of claims 58-60, wherein the one or more additional amino acid substitutions is selected from among T877A, W741C, W741L, W741R, L701H and H874Y.

62. The isolated AR polypeptide of any of claims 53-61, wherein the polypeptide comprises the sequence of amino acids set forth in SEQ ID NO: 5 or a variant that has at least or at least about 60%, 70%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or more sequence identity with the polypeptide having the sequence set forth in SEQ ID NO: 5, wherein the amino acid at position 876 is not phenylalanine.

63. The isolated AR polypeptide of any of claims 53-62 that is a recombinant protein.

64. An isolated nucleic acid molecule encoding the modified AR polypeptide of any of claims 53-62.

65. The isolated nucleic acid of claim 64, wherein the nucleic acid is a DNA or an RNA molecule.

66. The isolated nucleic acid of claim 64, wherein the nucleic acid is a cDNA molecule.

67. The isolated nucleic acid of claim 64 or claim 65, wherein the nucleic acid comprises the sequence of nucleic acids set forth in SEQ ID NO: 19 or a variant that has at least or at least about 60%, 70%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or more sequence identity with the polypeptide having the sequence set forth in SEQ ID NO: 19, wherein the nucleic acid codes encoding amino acid at position 876 does not encode phenylalanine.

68. The isolated nucleic acid of claim 67, wherein the nucleic acid molecule encodes leucine at position 876 of the AR polypeptide.

69. A vector, comprising the nucleic acid molecule of any of claim 64-68.

70. The vector of claim 69, wherein the vector is a viral or plasmid vector.

71. The vector of claim 69 or claim 70, wherein the nucleic acid is operably linked to a promoter.

72. The vector of claim 71, wherein the promoter is a constitutive or an inducible promoter.
73. A host cell, comprising the vector of any of claims 69-72.
74. The host cell of claim 73, wherein the cell is a prokaryotic cell or a eukaryotic cell.
75. The host cell of claim 74, wherein the cell is a mammalian cell, a bacterial cell, a yeast cell, an insect cell, a plant cell, or an amphibian cell.
76. A mutant AR polypeptide expressed by the host cell of any of claims 73-75.
77. A third-generation AR antagonist identified by the methods of any of claims 38-52.
78. A pharmaceutical composition comprising a third-generation AR antagonist identified by the methods of any of claims 38-52 and a pharmaceutically acceptable excipient.
79. A method of treatment comprising administering to a subject in need thereof a therapeutically effective amount of a pharmaceutical composition of claim 77 or claim 78, wherein the composition comprises a suitable pharmaceutical carrier.
80. The method of claim 79, wherein the subject has cancer.
81. The method of claim 80, wherein the cancer is a prostate cancer, a breast cancer, a bladder cancer or a hepatocellular cancer.
82. The method of claim 81, wherein the cancer is a castration resistant prostate cancer.
83. The method of any of claims 79-82, wherein the subject expresses a mutant AR.
84. The method of claim 83, wherein the mutant AR comprises a substitution or a deletion of the amino acid at amino acid position 876 in the AR polypeptide.
85. The method of claim 84, wherein the substitution is F876L.
86. The method of any of claims 71-85, wherein the third-generation AR antagonist is administered with an additional therapeutic agent.
87. The method of claim 86, wherein the third-generation AR antagonist and the additional therapeutic agent are administered sequentially, simultaneously or intermittently.
88. The method of claim 86 or claim 87, wherein the additional therapeutic agent is selected from among hormones, hormone receptor agonists or antagonists, corticosteroids, anti-emetic agents, analgesics, anti-cancer agents, anti-inflammatory agents, kinase inhibitors, HSP90 inhibitors, histone deacetylase (HDAC) inhibitors.
89. The method of claim 86 or claim 87, wherein the additional therapeutic agent is a gonadotropin-releasing hormone (GnRH) agonist or antagonist.

90. The method of claim 89, wherein the GnRH agonist is leuprolide, bruserelin or goserelin.
91. A microchip comprising the modified AR polypeptide of any of claims 53-62 or the nucleic acid of any of claims 64-68.
92. The microchip of claim 91, wherein the modified AR polypeptide comprises an amino acid substitution that is F876L or the nucleic acid encodes a modified AR polypeptide having an amino acid substitution that is F876L.
93. A kit comprising one or more reagents for the detection of nucleic acid encoding a modified AR polypeptide comprising a modification at amino acid position 876 or a modified AR polypeptide comprising a modification at amino acid position 876.
94. The kit of claim 93, wherein the modified AR polypeptide comprises an amino acid substitution that is F876L.
95. The kit of claim 93 or 94, comprising a pair oligonucleotide primers that flank the nucleic acid region encoding amino acid 876 of a AR polypeptide.
96. The kit of claim 93 or 94, comprising an oligonucleotide primer that
- (a) binds to nucleic acid encoding a modified AR that is modified at amino acid position 876; and
 - (b) does not bind to nucleic acid encoding the wild-type AR having phenylalanine at amino acid position 876.
97. The kit of claim 93 or 94, comprising a microchip comprising
- (a) a modified AR polypeptide having a modification that is F876S, or a portion thereof comprising a modification that is F876S; or
 - (b) a nucleic acid molecule encoding a mutant AR polypeptide having a modification that is F876S, or a portion thereof comprising a modification that is F876S.
98. A system for detecting a modified AR that is resistant to inhibition with an a first- or second-generation AR antagonist in a subject, comprising:
- (a) a sample containing a nucleic acid molecule encoding a AR polypeptide from the subject; and
 - (b) a microarray comprising nucleic acid encoding a mutant AR polypeptide or a portion thereof that is modified at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1.
99. The system of claim 98, wherein the microarray is contained on a microchip.
100. A system for detecting a modified AR that is resistant to inhibition with a first- or second- generation AR antagonist in a subject, comprising:

(a) a sample containing a nucleic acid molecule encoding a AR polypeptide from the subject; and

(b) a sequence specific nucleic acid probe, wherein the sequence specific nucleic acid probe:

(i) binds to nucleic acid encoding a modified AR that is modified at amino acid position 876; and

(ii) does not bind to nucleic acid encoding the wild-type AR having phenylalanine at amino acid position 876.

101. A system for detecting a modified AR that is resistant to inhibition with a first- or second- generation AR antagonist in a subject, comprising:

(a) a sample containing a nucleic acid molecule encoding a AR polypeptide from the subject; and

(b) a pair oligonucleotide primers that flank the nucleic acid region encoding amino acid 876 of a AR polypeptide.

102. An isolated antibody that binds to a modified AR polypeptide of any of claims 53-62, wherein the antibody does not bind to or binds with lower affinity to a wild-type AR polypeptide having the sequence of amino acids set forth in SEQ ID NO: 1.

103. The antibody of claim 102, wherein the modified AR polypeptide comprises an amino acid substitution that is F876L.

104. A method of maintenance therapy in a patient having a cancer, comprising:

(a) administering to the patient a maintenance therapy regimen comprising administering a therapeutically effective dose of first- or second- generation AR antagonist; and

(b) monitoring the patient at predetermined intervals of time over the course of the maintenance therapy regimen to determine whether the subject has mutation in an endogenous gene encoding AR that results in a modification at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1.

105. The method of claim 104, wherein monitoring comprises:

testing a sample containing a nucleic acid molecule encoding a AR polypeptide from the subject to determine whether the encoded AR polypeptide is modified at an amino acid position corresponding to amino acid position 876 of the amino acid sequence set forth in SEQ ID NO: 1.

106. The method of claim 104 or 105, wherein the method further comprises discontinuing maintenance therapy regimen if the subject has the mutation or continuing maintenance therapy regimen if the subject does not have the modification.

107. The method of claim 104 or 105, wherein the method further comprises administering third-generation AR antagonist that inhibits the modified AR if the subject has the modification.

108. The method of any of claims 104-107, wherein the modification in the AR polypeptide is F876L.

109. The method of any of claims 104-108, wherein the first- or second-generation AR antagonist inhibits a wild-type AR polypeptide by competitive antagonism.

110. The method of any of claims 104-109, wherein the second-generation AR antagonist is selected from among ARN-509, enzalutamide (MDV3100), and RD162.

111. The method any of claims 104-110, wherein the cancer is a prostate cancer, a breast cancer, a bladder cancer, or a hepatocellular cancer.

112. The method of claim 111, wherein the cancer is prostate cancer.

113. The method any claim 112, wherein the cancer is a castration resistant prostate cancer.

114. The method of any of claims 104-113, wherein the predetermined interval of time is every week, every month, every 2 months, every 3 months, every 4 months, every 5 months, every 6 months, every 7 months, every 8 months, every 9 months, every 10 months, every 11 months, or every year.

Claims:

1. A fusion protein comprising an androgen receptor (AR) polypeptide-comprising at least 95% sequence identity to a ligand binding domain having the amino acid sequence of amino acid positions 554 to 919 of SEQ ID NO: 1 and comprising an F876L amino acid substitution in SEQ ID NO: 1, wherein said AR polypeptide is fused to a heterologous polypeptide, a secretion signal sequence, an epitope tag, or an affinity tag.
2. The fusion protein of claim 1, wherein the AR polypeptide comprises the sequence of amino acids set forth in SEQ ID NO: 5.
3. The fusion protein of claim 1, wherein the AR polypeptide further comprises one or more amino acid substitutions in addition to the F876L amino acid substitution.
4. The fusion protein of claim 3, wherein the one or more additional amino acid substitutions is T877A, W741C, W741L, W741R, L701H or H874Y.
5. The fusion protein of claim 1, wherein the AR polypeptide further comprises at least one of:
 - (a) a substitution of the threonine at amino acid position 877 in SEQ ID NO: 1 by leucine, isoleucine, valine, alanine, phenylalanine, glycine, methionine, serine, cysteine, tryptophan, lysine, arginine, histidine, proline, tyrosine, asparagine, glutamine, aspartic acid or glutamic acid;
 - (b) a substitution of the tryptophan at amino acid position 741 in SEQ ID NO: 1 by leucine, isoleucine, valine, alanine, phenylalanine, glycine, methionine, serine, threonine, cysteine, lysine, arginine, histidine, proline, tyrosine, asparagine, glutamine, aspartic acid or glutamic acid;
 - (c) a substitution of the leucine at amino acid position 701 in SEQ ID NO: 1 by isoleucine, valine, alanine, phenylalanine, glycine, methionine, histidine, serine, threonine, cysteine, lysine, arginine, tryptophan, proline, tyrosine, asparagine, glutamine, aspartic acid, or glutamic acid; and
 - (d) a substitution of the histidine at amino acid position 874 in SEQ ID NO: 1 by leucine, isoleucine, valine, alanine, phenylalanine, glycine, methionine, serine, threonine,

cysteine, lysine, arginine, tryptophan, proline, tyrosine, asparagine, glutamine, aspartic acid or glutamic acid.

6. A cDNA molecule encoding an androgen receptor (AR) polypeptide comprising at least 95% sequence identity to a ligand binding domain having the amino acid sequence of amino acid positions 554 to 919 of SEQ ID NO:1 and comprising an F876L amino acid substitution in SEQ ID NO:1.

7. The cDNA molecule of claim 6, wherein the cDNA molecule comprises the nucleotide sequence set forth in SEQ ID NO: 19.

8. A vector, comprising the cDNA molecule of claim 6.

9. The vector of claim 8, wherein the vector is a viral or plasmid vector.

10. The vector of claim 8, wherein the cDNA molecule is operably linked to a promoter.

11. The vector of claim 10, wherein the promoter is a constitutive or an inducible promoter.

12. An isolated host cell, comprising the vector of claim 8.

13. The isolated host cell of claim 12, wherein the isolated host cell is a prokaryotic cell.

14. The isolated host cell of claim 13, wherein the isolated host cell is a bacterial cell.

15. A microchip comprising the fusion protein of claim 1 or the cDNA molecule of claim 6.

16. A system for detecting a modified AR that is resistant to inhibition with a first- or second-generation AR antagonist in a subject, comprising:

(a) a sample containing a nucleic acid molecule encoding an AR polypeptide from the subject; and

(b) a microarray comprising the cDNA molecule of claim 6.

17. The system of claim 16, wherein the microarray is contained on a microchip.

18. The fusion protein of claim 1, wherein the heterologous polypeptide is a heterologous DNA binding domain, a fluorescent protein, or a bioluminescent protein.

19. The fusion protein of claim 18, wherein the heterologous DNA binding domain is selected from the group consisting of GAL4 and LexA.
20. The fusion protein of claim 18, wherein the AR polypeptide is linked to the heterologous DNA binding domain via a peptide linker.
21. The fusion protein of claim 18, wherein the fluorescent protein is selected from the group consisting of green fluorescent protein (GFP), red fluorescent protein (RFP), cyan fluorescent protein (CFP), yellow fluorescence protein (YFP), and blue fluorescent protein (BFP).
22. The fusion protein of claim 1, wherein the epitope tag is selected from the group consisting of c-myc, V-5, hemagglutinin (HA), and FLAG.
23. The fusion protein of claim 1, wherein the affinity tag is selected from the group consisting of biotin, strep-tag, chitin binding protein (CBP), maltose binding protein (MBP), glutathione-S-transferase (GST), and a poly(His) tag.
24. The isolated host cell of claim 12, wherein the isolated host cell is a eukaryotic cell.
25. The isolated host cell of claim 24, wherein the isolated host cell is a mammalian cell, a yeast cell, an insect cell, a plant cell, or an amphibian cell.
26. The fusion protein of claim 1, wherein the AR polypeptide has at least 96% sequence identity to the ligand binding domain.
27. The fusion protein of claim 1, wherein the AR polypeptide has at least 97% sequence identity to the ligand binding domain.
28. The fusion protein of claim 1, wherein the AR polypeptide has at least 98% sequence identity to the ligand binding domain.
29. The cDNA molecule of claim 6, wherein the AR polypeptide has at least 96% sequence identity to the ligand binding domain.
30. The cDNA molecule of claim 6, wherein the AR polypeptide has at least 97% sequence identity to the ligand binding domain.

31. The cDNA molecule of claim 6, wherein the AR polypeptide has at least 98% sequence identity to the ligand binding domain.