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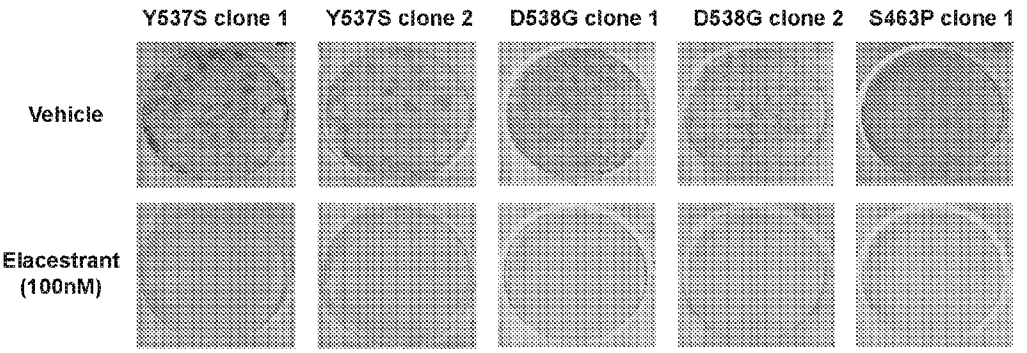

FIG. 1

(57) Abrégé/Abstract:
Disclosed herein are methods of treating a drug resistant estrogen receptor alpha¬ positive cancer in a subject having a mutant estrogen receptor alpha, the method comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof, wherein the mutant estrogen receptor alpha comprises one or more mutations selected from the group consisting of D538G, $Y537X_1$, $L536X_2$, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: Xi is S, N, or C; and X_2 is R or Q. In some embodiments, the drug resistant estrogen receptor alpha-positive cancer is selected from the group consisting of breast cancer, uterine cancer, ovarian cancer, and pituitary cancer.

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(54) Title: METHODS FOR TREATING CANCER IN MODELS HARBORING ESR1 MUTATIONS

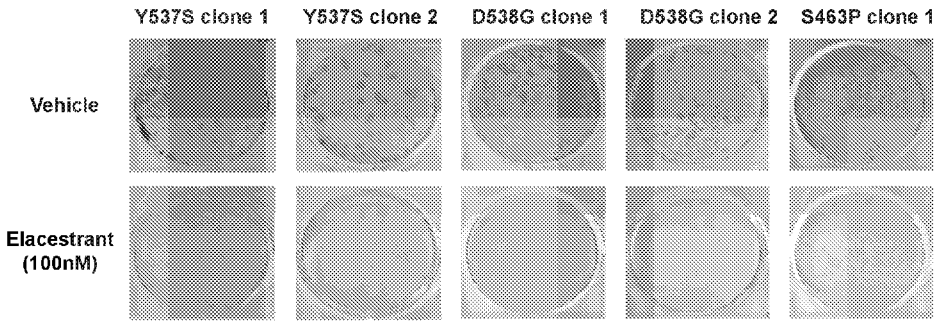

FIG. 1

(57) Abstract: Disclosed herein are methods of treating a drug resistant estrogen receptor alpha¬ positive cancer in a subject having a mutant estrogen receptor alpha, the method comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof, wherein the mutant estrogen receptor alpha comprises one or more mutations selected from the group consisting of D538G, $Y537X_1$, $L536X_2$, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: Xi is S, N, or C; and X_2 is R or Q. In some embodiments, the drug resistant estrogen receptor alpha-positive cancer is selected from the group consisting of breast cancer, uterine cancer, ovarian cancer, and pituitary cancer.

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[Continued on next page]

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METHODS FOR TREATING CANCER IN MODELS HARBORING ESR1 MUTATIONS

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority under 35 U.S.C. § 119(e) to United States Provisional Patent Application No. 62/776,338, filed December 6, 2018. The entire contents of the aforementioned application are hereby incorporated by reference in its entirety, including drawings.

TECHNICAL FIELD

[0002] The present disclosure provides methods of providing anti-tumor activity using elacestrant in cancer models harboring ESR1 mutations resistant to standard of care therapies. The present disclosure also relates to methods of treating estrogen positive (ER+) cancers having ESR1 mutations that can contribute to endocrine resistance where the cancer is effectively treated using elacestrant.

BACKGROUND

[0003] Breast cancer is divided into three subtypes based on expression of three receptors: estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor-2 (Her2). Overexpression of ERs is found in many breast cancer patients. ER-positive (ER+) breast cancers comprise two-thirds of all breast cancers. Other than breast cancer, estrogen and ERs are associated with, for example, ovarian cancer, colon cancer, prostate cancer and endometrial cancer.

[0004] ERs can be activated by estrogen and translocate into the nucleus to bind to DNA, thereby regulating the activity of various genes. See, e.g., Marino et al., "Estrogen Signaling Multiple Pathways to Impact Gene Transcription," *Curr. Genomics* 7(8): 497-508 (2006); and Heldring et al., "Estrogen Receptors: How Do They Signal and What Are Their Targets," *Physiol. Rev.* 87(3): 905-931 (2007).

[0005] Agents that inhibit estrogen production, such as aromatase inhibitors (AIs, e.g., letrozole, anastrozole and exemestane), or those that directly block ER activity, such as selective estrogen receptor modulators (SERMs, e.g., tamoxifen, toremifene, droloxifene, idoxifene, raloxifene, lasofoxifene, arzoxifene, miproxifene, levormeloxifene, and EM-652 (SCH 57068)) and selective estrogen receptor degraders (SERDs, e.g., fulvestrant, TAS-108 (SR16234), ZK191703, RU58668, GDC-0810 (ARN-810), GW5638/DPC974, SRN-927,

ICI182780 and AZD9496), have been used previously or are being developed in the treatment of ER-positive breast cancers.

[0006] SERMs and AIs are often used as a first-line adjuvant systemic therapy for ER-positive breast cancer. AIs suppress estrogen production in peripheral tissues by blocking the activity of aromatase, which turns androgen into estrogen in the body. However, AIs cannot stop the ovaries from making estrogen. Thus, AIs are mainly used to treat post-menopausal women. Furthermore, as AIs are more effective than the SERM tamoxifen with fewer serious side effects, AIs may also be used to treat pre-menopausal women with their ovarian function suppressed. See, e.g., Francis et al., "Adjuvant Ovarian Suppression in Premenopausal Breast Cancer," the *N. Engl. J. Med.*, 372:436-446 (2015).

[0007] Resistance to endocrine therapy is a challenging aspect in the management of patients with estrogen receptor positive (ER+) breast cancer. Recent studies have demonstrated that acquired resistance can develop after treatment with aromatase inhibitors through the emergence of mutations in the estrogen receptor 1 (ESR1) gene. While initial treatment with these agents may be successful, many patients eventually relapse with drug-resistant breast cancers. Mutations affecting the ER have emerged as one potential mechanism for the development of this resistance. See, e.g., Robinson et al., "Activating ESR1 mutations in hormone-resistant metastatic breast cancer," *Nat Genet.* 45:1446–51 (2013). Mutations in the ligand-binding domain (LBD) of ER are found in 20-40% of metastatic ER-positive breast tumor samples from patients who received at least one line of endocrine treatment. Jeselsohn, et al., "ESR1 mutations—a mechanism for acquired endocrine resistance in breast cancer," *Nat. Rev. Clin. Oncol.*, 12:573-83 (2015).

[0008] Therefore, there remains a need for more durable and effective ER-targeted therapies to overcome some of the challenges associated with current endocrine therapies and to combat the development of resistance.

SUMMARY OF THE INVENTION

[0009] In one aspect, the disclosure relates to a method of inhibiting and degrading a mutant estrogen receptor alpha positive cancer in a subject comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof.

[0010] Embodiments of this aspect of the invention may include one or more of the following optional features. In some embodiments, the mutant estrogen receptor alpha positive cancer comprises one or more mutations selected from the group consisting of

D538G, Y537X_1, L536X_2, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: X_1 is S, N, or C; and X_2 is R or Q. In some embodiments, the mutation is Y537S. In some embodiments, the mutation is D538G. In some embodiments, the mutant estrogen receptor alpha positive cancer is resistant to a drug selected from the group consisting of anti-estrogens, aromatase inhibitors, and combinations thereof. In some embodiments, the mutant estrogen receptor alpha positive cancer is selected from the group consisting of breast cancer, uterine cancer, ovarian cancer, and pituitary cancer. In some embodiments, the mutant estrogen receptor alpha positive cancer is advanced or metastatic breast cancer. In some embodiments, the mutant estrogen receptor alpha positive cancer is breast cancer. In some embodiments, the subject is a post-menopausal woman. In some embodiments, the subject is a pre-menopausal woman. In some embodiments, the subject is a post-menopausal woman who had relapsed or progressed after previous treatment with selective estrogen receptor modulators (SERMs) and/or aromatase inhibitors (AIs). In some embodiments, the elacestrant is administered to the subject at a dose of from about 200 mg/day to about 500 mg/day. In some embodiments, the elacestrant is administered to the subject at a dose of about 200 mg/day, about 300 mg/day, about 400 mg/day, or about 500 mg/day. In some embodiments, the elacestrant is administered to the subject at a dose that is the maximum tolerated dose for the subject. In some embodiments, the method further comprises identifying the subject for treatment by measuring increased expression of one or more genes selected from ABL1, AKT1, AKT2, ALK, APC, AR, ARID1A, ASXL1, ATM, AURKA, BAP, BAP1, BCL2L11, BCR, BRAF, BRCA1, BRCA2, CCND1, CCND2, CCND3, CCNE1, CDH1, CDK4, CDK6, CDK8, CDKN1A, CDKN1B, CDKN2A, CDKN2B, CEBPA, CTNNB1, DDR2, DNMT3A, E2F3, EGFR, EML4, EPHB2, ERBB2, ERBB3, ESR1, EWSR1, FBXW7, FGF4, FGFR1, FGFR2, FGFR3, FLT3, FRS2, HIF1A, HRAS, IDH1, IDH2, IGF1R, JAK2, KDM6A, KDR, KIF5B, KIT, KRAS, LRP1B, MAP2K1, MAP2K4, MCL1, MDM2, MDM4, MET, MGMT, MLL, MPL, MSH6, MTOR, MYC, NF1, NF2, NKX2-1, NOTCH1, NPM, NRAS, PDGFRA, PIK3CA, PIK3R1, PML, PTEN, PTPRD, RARA, RB1, RET, RICTOR, ROS1, RPTOR, RUNX1, SMAD4, SMARCA4, SOX2, STK11, TET2, TP53, TSC1, TSC2, and VHL. In some embodiments, the one or more genes is selected from AKT1, AKT2, BRAF, CDK4, CDK6, PIK3CA, PIK3R1, and MTOR. In some embodiments, the ratio of the concentration of elacestrant or a salt or solvate thereof in the tumor to the concentration of elacestrant or a salt or solvate thereof in plasma (T/P) following administration is at least about 15.

[0011] In another aspect, the disclosure relates to a method of treating a drug resistant estrogen receptor alpha-positive cancer in a subject having a mutant estrogen receptor alpha, the method comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof, wherein the mutant estrogen receptor alpha comprises one or more mutations selected from the group consisting of D538G, $Y537X_1$, $L536X_2$, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: X_1 is S, N, or C; and X_2 is R or Q.

[0012] Embodiments of this aspect of the invention may include one or more of the following optional features. In some embodiments, the cancer is resistant to a drug selected from the group consisting of anti-estrogens, aromatase inhibitors, and combinations thereof. In some embodiments, the anti-estrogens are selected from the group consisting of tamoxifen, toremifene and fulvestrant and the aromatase inhibitors are selected from the group consisting of exemestane, letrozole and anastrozole. In some embodiments, the drug resistant estrogen receptor alpha-positive cancer is selected from the group consisting of breast cancer, uterine cancer, ovarian cancer, and pituitary cancer. In some embodiments, the cancer is advanced or metastatic breast cancer. In some embodiments, the cancer is breast cancer. In some embodiments, the subject is a post-menopausal woman. In some embodiments, the subject is a pre-menopausal woman. In some embodiments, the subject is a post-menopausal woman who had relapsed or progressed after previous treatment with SERMs and/or AIs. In some embodiments, the subject expresses at least one mutant estrogen receptor alpha selected from the group consisting of D538G, Y537S, Y537N, Y537C, E380Q, S463P, L536R, L536Q, P535H, V392I and V534E. In some embodiments, the mutation includes Y537S. In some embodiments, the mutation includes D538G. In some embodiments, the method further comprises identifying the subject for treatment by measuring increased expression of one or more genes selected from ABL1, AKT1, AKT2, ALK, APC, AR, ARID1A, ASXL1, ATM, AURKA, BAP, BAP1, BCL2L11, BCR, BRAF, BRCA1, BRCA2, CCND1, CCND2, CCND3, CCNE1, CDH1, CDK4, CDK6, CDK8, CDKN1A, CDKN1B, CDKN2A, CDKN2B, CEBPA, CTNNB1, DDR2, DNMT3A, E2F3, EGFR, EML4, EPHB2, ERBB2, ERBB3, ESR1, EWSR1, FBXW7, FGF4, FGFR1, FGFR2, FGFR3, FLT3, FRS2, HIF1A, HRAS, IDH1, IDH2, IGF1R, JAK2, KDM6A, KDR, KIF5B, KIT, KRAS, LRP1B, MAP2K1, MAP2K4, MCL1, MDM2, MDM4, MET, MGMT, MLL, MPL, MSH6, MTOR, MYC, NF1, NF2, NKX2-1, NOTCH1, NPM, NRAS, PDGFRA, PIK3CA, PIK3R1, PML, PTEN, PTPRD, RARA, RB1, RET, RICTOR, ROS1, RPTOR, RUNX1, SMAD4, SMARCA4, SOX2, STK11, TET2, TP53, TSC1, TSC2, and VHL. In some embodiments,

the one or more genes is selected from AKT1, AKT2, BRAF, CDK4, CDK6, PIK3CA, PIK3R1, and MTOR. In some embodiments, the elacestrant is administered to the subject at a dose of from about 200 to about 500 mg/day. In some embodiments, the elacestrant is administered to the subject at a dose of about 200 mg, about 300 mg, about 400 mg, or about 500 mg. In some embodiments, the elacestrant is administered to the subject at a dose of about 300 mg/day. In some embodiments, the ratio of the concentration of elacestrant or a salt or solvate thereof in the tumor to the concentration of elacestrant or a salt or solvate thereof in plasma (T/P) following administration is at least about 15.

[0013] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Methods and materials are described herein for use in the present invention; other suitable methods and materials known in the art can also be used. The materials, methods, and examples are illustrative only and not intended to be limiting. All publications, patent applications, patents, sequences, database entries, and other references mentioned herein are incorporated by reference in their entirety. In case of conflict, the present specification, including definitions, will control.

[0014] Other features and advantages of the invention will be apparent from the following detailed description and figures, and from the claims.

BRIEF DESCRIPTION OF THE FIGURES

[0015] The following figures are provided by way of example and are not intended to limit the scope of the claimed invention.

[0016] FIG. 1. The representative pictures presented in the top row visualize tumor cells treated with vehicle control for the Y537S clone 1, Y537S clone 2, D538G clone 1, D538G clone 2, and S463P clone 1 mutated cancer cell lines. The pictures presented in the bottom row visualize the Y537S clone 1, Y537S clone 2, D538G clone 1, D538G clone 2, and S463P clone 1 tumor cells treated with elacestrant at 100 nM.

[0017] FIG. 2. Mean +/- SEM tumor volumes over time in mouse xenograft models treated with vehicle control, elacestrant (30, 60, and 120 mg/kg) and fulvestrant (1 mg/dose).

[0018] FIG. 3A. Fold change relative to control of progesterone receptor (PgR) for tumor cell models having wild type, S463P, D538G, and Y537S mutations treated with vehicle control, elacestrant (10, 100, and 1000 nM), E2 (10pM), and fulvestrant (10, 100, 1000 nM).

[0019] FIG. 3B. Fold change relative to control of growth regulated by estrogen (GREB1) in tumor cell models having wild type, S463P, D538G, and Y537S mutations treated with

vehicle control, elacestrant (10, 100, and 1000 nM), E2 (10pM), and fulvestrant (10, 100, 1000 nM).

[0020] FIG. 3C. Fold change relative to control of trefoil factor 1 (TFF1) in tumor cell models having wild type, S463P, D538G, and Y537S mutations treated with vehicle control, elacestrant (10, 100, and 1000 nM), E2 (10pM), and fulvestrant (10, 100, 1000 nM).

[0021] FIG. 4A. Mean +/- SEM tumor volumes over time in athymic nude mice implanted with ST2535-HI PDX xenograft (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation treated with vehicle control and elacestrant (30 and 60 mg/kg).

[0022] FIG. 4B. Mean +/- SEM tumor volumes over time in athymic nude mice implanted with CTG-1211-HI PDX xenograft (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose).

[0023] FIG. 4C. Mean +/- SEM tumor volumes over time in athymic nude mice implanted with WHIM43-HI PDX xenograft (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg) and fulvestrant (3 mg/dose).

[0024] FIG. 5A. Fold change over vehicle control of progesterone receptor (PgR) mRNA levels in the ST2535-HI PDX xenograft model (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation treated with vehicle control and elacestrant (30 and 60 mg/kg).

[0025] FIG. 5B. A Western blot illustrating PgR expression in the ST2535-HI PDX xenograft model with an ESR1:D538G mutation treated with vehicle control and elacestrant (30 and 60 mg/kg).

[0026] FIG. 5C. Fold change over vehicle control of progesterone receptor (PgR) mRNA levels in the CTG-1211-HI PDX xenograft model (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose).

[0027] FIG. 5D. A Western blot illustrating PgR expression in the CTG-1211-HI PDX xenograft model with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant.

[0028] FIG. 5E. Fold change over vehicle control of progesterone receptor (PgR) mRNA levels in the WHIM43-HI PDX xenograft model (previously treated with tamoxifen,

aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose).

[0029] FIG. 5F. A Western blot illustrating PgR expression in the WHIM43-HI PDX xenograft model with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant.

[0030] FIG. 6A. Mean +/- SEM tumor volumes over time in the ST941-HI PDX model harboring an ESR1:Y537S mutation treated with vehicle control, elacestrant (10, 30, and 60 mg/kg) fulvestrant (3 mg/dose), serd1 dose 1, and serd1 dose 2.

[0031] FIG. 6B. Mean +/- SEM tumor volumes over time in the ST941-HI PDX model harboring an ESR1:Y537S mutation treated with vehicle control, elacestrant (10, 30, and 60 mg/kg) fulvestrant (3 mg/dose), serd2 dose 1, and serd2 dose 2.

[0032] FIG. 7A. Fold change over vehicle control relative to progesterone receptor (PgR) mRNA levels in the ST941-HI PDX model harboring an ESR1:Y537S mutation treated with vehicle control, fulvestrant (3 mg/dose), elacestrant (30 mg/kg), serd1 dose 1, serd1 dose 2, serd2 dose 1, and serd2 dose 2.

[0033] FIG. 7B. A Western blot illustrating the ST941-HI PDX model harboring an ESR1:Y537S mutation demonstrating PgR expression treated with vehicle control, fulvestrant (3 mg/kg), elacestrant (30 mg/kg), serd1 dose 1, and serd1 dose 2.

[0034] FIG. 7C. A Western blot illustrating the ST941-HI PDX model harboring an ESR1:Y537S mutation demonstrating PgR expression treated with vehicle control, fulvestrant (3 mg/kg), elacestrant (30 mg/kg), serd2 dose 1, and serd2 dose 2.

[0035] FIG. 8A. *In vitro* cell viability (% of control) provided with respect to Log[Concentration (μm)] for the ST941-HI PDX cell line.

[0036] FIG. 8B. Fold change over vehicle control of progesterone receptor (PgR) mRNA levels plotted with respect to the concentration of elacestrant (0, 10, 100, and 1000 nM) and fulvestrant (0, 10, 100, and 1000 nM) used in treating *in vitro* ST941-HI cell line derived from PDX.

[0037] FIG. 9A. Mean +/- SEM tumor volumes in mice implanted with the ST941-HI PDX harboring an ESR1:Y537S mutation plotted with respect to time and their treatment with vehicle control, elacestrant (10, 30, and 60 mg/kg) and fulvestrant (3 mg/dose).

[0038] FIG. 9B. Fold change relative to control of progesterone receptor (PgR) mRNA expression in the ST941-HI PDX model plotted with respect to their treatment with vehicle control, fulvestrant (3 mg/dose), and elacestrant (10, 30, and 60 mg/kg).

[0039] FIG. 10A. Mean +/- SEM tumor volumes over time in mice implanted with the WHIM20 PDX xenograft with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30, and 60 mg/kg) and fulvestrant (3 mg/dose).

[0040] FIG. 10B. Fold change relative to vehicle control of progesterone receptor (PgR) in the WHIM20 PDX xenograft model with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose).

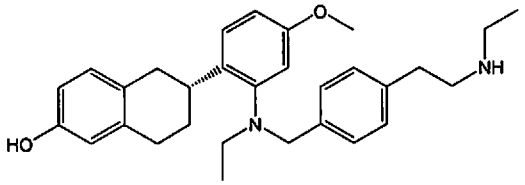
[0041] FIG. 10C. Fold change relative to control of trefoil factor 1 (TFF1) in the WHIM20 PDX xenograft model with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose).

[0042] FIG. 10D. Fold change relative to control of growth regulated by estrogen (GREB1) in the WHIM20 PDX xenograft model with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose).

[0043] FIG. 10E. A Western blot illustrating the WHIM20 PDX xenograft model with an ESR1:Y537S^{hom} mutation showing PgR expression and treated with vehicle control, fulvestrant (3 mg/dose), and elacestrant (30 and 60 mg/kg).

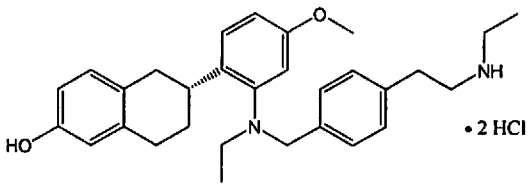
DETAILED DESCRIPTION OF THE INVENTION

[0044] As used herein, elacestrant or "RAD1901" is an orally bioavailable selective estrogen receptor degrader (SERD) and has the following chemical structure:



Elacestrant

including salts, solvates (e.g. hydrate), and prodrugs thereof. Preclinical data have demonstrated elacestrant is effective in inhibiting tumor growth in models of ER+ breast cancer with both wild-type and mutant ESR1. In some embodiments described herein, elacestrant is administered as the bis-hydrochloride (•2HCl) salt having the following chemical structure:



Elacestrant dihydrochloride.

[0045] In postmenopausal women, the current standard of care for ER+ cancers, such as breast cancer, involves inhibiting the ER pathway by: 1) inhibiting the synthesis of estrogen (aromatase inhibitors (AI)); 2) directly binding to ER and modulating its activity using SERMs (e.g., tamoxifen); and/or 3) directly binding to ER and causing receptor degradation using SERDs (e.g., fulvestrant). In premenopausal women, the current standard of care would additionally include ovarian suppression through oophorectomy or a luteinizing hormone-releasing hormone (LHRH) agonist. Despite patients responding typically well to clinically approved treatments, ESR1 mutations can frequently occur (e.g., 20-40%) in metastatic breast cancer and can contribute to endocrine resistance. While the response of ESR1 mutations to AIs and/or SERDs is not fully understood, recent data from the ctDNA analysis of the PALOMA-3 trial of palbociclib and fulvestrant versus placebo and fulvestrant demonstrated a trend towards selection of the ESR1 Y537S mutation after fulvestrant treatment. This data shows the trend of ESR1s to mutate, in combination with the requirement to dose fulvestrant intramuscularly, and highlights the need for new and/or improved orally-bioavailable endocrine therapies that have efficacy against ESR1 and all ESR1 mutations.

[0046] The unexpected efficacy of elacestrant to target tumors hardly responsive to fulvestrant treatments and in tumors expressing mutant ERα may be due to the unique interactions between elacestrant and ERα. Structural models of ERα bound to elacestrant and other ERα-binding compounds were analyzed to obtain information about the specific binding interactions. Computer modeling showed that elacestrant-ERα interactions are not likely to be affected by mutants of LBD of ERα, e.g., Y537X mutant wherein X was S, N, or C; D538G; and S463P, which account for about 81.7% of LBD mutations found in a recent study of metastatic ER positive breast tumor samples from patients who received at least one line of endocrine treatment. This resulted in identification of specific residues in the C-terminal ligand-binding domains of ERα that are critical to binding, information that can be used to develop compounds that bind and antagonize not only wild-type ERα but also certain mutations and variants thereof.

[0047] Based on these results, methods are provided herein for inhibiting growth or producing regression of an ERα positive cancer or tumor in a subject in need thereof by administering to the subject a therapeutically effective amount of elacestrant or a solvate (e.g., hydrate) or salt thereof. In certain embodiments, administration of elacestrant or a salt or solvate (e.g., hydrate) thereof has additional therapeutic benefits in addition to inhibiting tumor growth, including for example inhibiting cancer cell proliferation or inhibiting ERα activity (e.g., by inhibiting estradiol binding or by degrading ERα). In certain embodiments, the method does not provide negative effects to muscles, bones, breast, and uterus.

[0048] In certain embodiments of the tumor growth inhibition or regression methods provided herein, the methods further comprise a step of determining whether a patient has a tumor expressing ERα prior to administering elacestrant or a solvate (e.g., hydrate) or salt thereof. In certain embodiments of the tumor growth inhibition or regression methods provided herein, the methods further comprise a step of determining whether the patient has a tumor expressing mutant ERα prior to administering elacestrant or a solvate (e.g., hydrate) or salt thereof. In certain embodiments of the tumor growth inhibition or regression methods provided herein, the methods further comprise a step of determining whether a patient has a tumor expressing ERα that is responsive or non-responsive to an AI, a SERD (e.g., fulvestrant), and/or a SERM (e.g. tamoxifen) treatment prior to administering elacestrant or a solvate (e.g., hydrate) or salt thereof. These determinations may be made using any method of expression detection known in the art and may be performed *in vitro* using a tumor or tissue sample removed from the subject.

[0049] In the methods described herein, elacestrant is demonstrated to inhibit the growth of several PDX models harboring the ESR1:D538G and ESR1:Y537S mutations, including models that are palbociclib-resistant, fulvestrant-resistant, and have been previously treated with aromatase inhibitors/tamoxifen/fulvestrant. Elacestrant is also demonstrated to both degrade ERs and inhibit ER signaling in PDX models harboring the ESR1:D538G mutation. Elacestrant is efficacious in the *in vitro* and the *in vivo* ST941-HI model that harbors a Y537S mutation. Additionally, two SERDs having acrylic acid side chains demonstrated partial growth inhibition *in vivo* in the ST941-HI PDX model. Fulvestrant, while being efficacious *in vitro,* demonstrated a lack of activity *in vivo* in the ST941-HI PDX model. While elacestrant and fulvestrant both demonstrate partial efficacy in the WHIM20 model harboring an ESR1:Y537S mutation despite both agents degrading ER and inhibiting ER signaling, elacestrant's role and combination with inhibitors of other oncogenic drivers in tumor growth is providing advances in tumor treatments with improved efficacy.

[0050] The role of ESR1 mutations in endocrine resistance and their impact on the efficacy of endocrine treatments is a complicated relationship. Indeed, recent data from the ctDNA analysis of the PALOMA3 trial of palbociclib and fulvestrant versus placebo and fulvestrant demonstrated a trend towards selection of the ESR1:Y537S mutation after fulvestrant treatment. This research suggests there may be certain contexts of ESR1 mutations where fulvestrant may have limited activity. Therefore, the studies herein indicating the effective use of elacestrant as a treatment for cancers having efficacy against all ESR1 mutations is a promising discovery.

Definitions

[0051] As used herein, the following definitions shall apply unless otherwise indicated.

[0052] As used herein, the terms "RAD1901" and "elacestrant" refer to the same chemical compound and are used interchangeably.

[0053] "Inhibiting growth" of an ERα-positive tumor as used herein may refer to slowing the rate of tumor growth, or halting tumor growth entirely.

[0054] "Tumor regression" or "regression" of an ERα-positive tumor as used herein may refer to reducing the maximum size of a tumor. In certain embodiments, administration of a combination as described herein, or solvates (e.g., hydrate) or salts thereof may result in a decrease in tumor size versus baseline (i.e., size prior to initiation of treatment), or even eradication or partial eradication of a tumor. Accordingly, in certain embodiments the methods of tumor regression provided herein may be alternatively characterized as methods of reducing tumor size versus baseline.

[0055] "Tumor" as used herein is a malignant tumor, and is used interchangeably with "cancer."

[0056] "Estrogen receptor alpha" or "ERα" as used herein refers to a polypeptide comprising, consisting of, or consisting essentially of the wild-type ERα amino acid sequence, which is encoded by the gene ESR1.

[0057] A tumor that is "positive for estrogen receptor alpha," "ERα-positive," "ER+," or "ERα+" as used herein refers to a tumor in which one or more cells express at least one isoform of ERα.

[0058] "Standard of Care Therapies" as used herein refers to agents known and commonly used to treat cancers such as breast cancer including aromatase inhibitors (AIs, e.g., letrozole, anastrozole and exemestane), selective estrogen receptor modulators (SERMs, e.g., tamoxifen, toremifene, droloxifene, idoxifene, raloxifene, lasofoxifene, arzoxifene, miproxifene, levormeloxifene, and EM-652 (SCH 57068)), and/or selective estrogen receptor

degraders (SERDs, e.g., fulvestrant, TAS-108 (SR16234), ZK191703, RU58668, GDC-0810 (ARN-810), GW5638/DPC974, SRN-927, ICI182782 and AZD9496).

Methods of Treatment

[0059] In some embodiments, the disclosure relates to a method of inhibiting and degrading a mutant estrogen receptor alpha positive cancer in a subject comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof.

[0060] In other embodiments, the disclosure relates to a method of treating a drug resistant estrogen receptor alpha-positive cancer in a subject having a mutant estrogen receptor alpha, the method comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof, wherein the mutant estrogen receptor alpha comprises one or more mutations selected from the group consisting of D538G, $Y537X_1$, $L536X_2$, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: X_1 is S, N, or C; and X_2 is R or Q.

Administration of Elacestrant

[0061] Elacestrant or solvates (e.g., hydrate) or salts thereof, when administered to a subject, have a therapeutic effect on one or more cancers or tumors. Tumor growth inhibition or regression may be localized to a single tumor or to a set of tumors within a specific tissue or organ, or may be systemic (i.e., affecting tumors in all tissues or organs).

[0062] As elacestrant is known to preferentially bind ERα versus estrogen receptor beta (ERβ), unless specified otherwise, estrogen receptor, estrogen receptor alpha, ERα, ER, and wild-type ERα are used interchangeably herein. In certain embodiments, ER+ cells overexpress ERα. In certain embodiments, the patient has one or more cells within the tumor expressing one or more forms of ERβ. In certain embodiments, the ERα-positive tumor and/or cancer is associated with breast, uterine, ovarian, or pituitary cancer. In certain of these embodiments, the patient has a tumor located in breast, uterine, ovarian, or pituitary tissue. In those embodiments where the patient has a tumor located in the breast, the tumor may be associated with luminal breast cancer that may or may not be positive for HER2, and for HER2+ tumors, the tumors may express high or low HER2. In other embodiments, the patient has a tumor located in another tissue or organ (e.g., bone, muscle, brain), but is nonetheless associated with breast, uterine, ovarian, or pituitary cancer (e.g., tumors derived from migration or metastasis of breast, uterine, ovarian, or pituitary cancer). Accordingly, in certain embodiments of the tumor growth inhibition or tumor regression methods provided herein, the tumor being targeted is a metastatic tumor and/or the tumor has an overexpression

of ER in other organs (e.g., bones and/or muscles). In certain embodiments, the tumor being targeted is a brain tumor and/or cancer. In certain embodiments, the tumor being targeted can be more sensitive to a treatment of elacestrant than treatment with another SERD (e.g., fulvestrant, TAS-108 (SR16234), ZK191703, RU58668, GDC-0810 (ARN-810), GW5638/DPC974, SRN-927, and AZD9496), Her2 inhibitors (e.g., trastuzumab, lapatinib, ado-trastuzumab emtansine, and/or pertuzumab), chemo therapy (e.g., abraxane, adriamycin, carboplatin, cytoxan, daunorubicin, doxil, ellence, fluorouracil, gemzar, helaven, Ixempra, methotrexate, mitomycin, micoxantrone, navelbine, taxol, taxotere, thiotepa, vincristine, and xeloda), aromatase inhibitor (e.g., anastrozole, exemestane, and letrozole), selective estrogen receptor modulators (e.g., tamoxifen, raloxifene, lasofoxifene, and/or toremifene), angiogenesis inhibitor (e.g., bevacizumab), and/or rituximab.

[0063] In addition to demonstrating the ability of elacestrant to inhibit tumor growth in tumors expressing wild-type ERα, elacestrant exhibits the ability to inhibit the growth of tumors expressing a mutant form of ERα, namely Y537S ERα. Computer modeling evaluations of examples of ERα mutations showed that none of these mutations were expected to impact the LBD or specifically hinder elacestrant binding, e.g., ERα having one or more mutants selected from the group consisting of ERα with Y537X mutant wherein X is S, N, or C, ERα with D538G mutant, and ERα with S463P mutant. Based on these results, methods are provided herein for inhibiting growth or producing regression of a tumor that is positive for ERα having one or more mutants within the ligand-binding domain (LBD), selected from the group consisting of Y537X1 wherein X1 is S, N, or C, D538G, L536X2 wherein X2 is R or Q, P535H, V534E, S463P, V392I, E380Q, especially Y537S ERα, in a subject with cancer by administering to the subject a therapeutically effective amount of elacestrant or solvates (e.g., hydrate) or salts thereof. "Mutant ERα" as used herein refers to ERα comprising one or more substitutions or deletions, and variants thereof comprising, consisting of, or consisting essentially of an amino acid sequence with at least 80%, at least 85%, at least 90%, at least 95%, at least 97%, at least 98%, at least 99%, or at least 99.5% identity to the amino acid sequence of ERα.

[0064] In addition to inhibiting breast cancer tumor growth in an animal xenograft model, elacestrant exhibits significant accumulation within tumor cells, and is capable of penetrating the blood-brain barrier. The ability to penetrate the blood-brain barrier was confirmed by showing that elacestrant administration significantly prolonged survival in a brain metastasis xenograft model. Accordingly, in certain embodiments of the tumor growth inhibition or tumor regression methods provided herein, the ERα-positive tumor being targeted is located

in the brain or elsewhere in the central nervous system. In certain of these embodiments, the ERα-positive tumor is primarily associated with brain cancer. In other embodiments, the ERα-positive tumor is a metastatic tumor that is primarily associated with another type of cancer, such as breast, uterine, ovarian, or pituitary cancer, or a tumor that has migrated from another tissue or organ. In certain of these embodiments, the tumor is a brain metastases, such as breast cancer brain metastases (BCBM). In certain embodiments of the methods disclosed herein, elacestrant or solvates (e.g., hydrate) or salts thereof accumulate in one or more cells within a target tumor.

[0065] In certain embodiments of the methods disclosed herein, elacestrant or solvates (e.g., hydrate) or salts thereof preferably accumulate in tumor at a T/P (elacestrant concentration in tumor/elacestrant concentration in plasma) ratio of about 15 or higher, about 18 or higher, about 19 or higher, about 20 or higher, about 25 or higher, about 28 or higher, about 30 or higher, about 33 or higher, about 35 or higher, or about 40 or higher.

Dosage

[0066] A therapeutically effective amount of a combination of elacestrant or solvates (e.g., hydrate) or salts thereof for use in the methods disclosed herein is an amount that, when administered over a particular time interval, results in achievement of one or more therapeutic benchmarks (e.g., slowing or halting of tumor growth, resulting in tumor regression, cessation of symptoms, etc.). The combination for use in the presently disclosed methods may be administered to a subject one time or multiple times. In those embodiments wherein the compounds are administered multiple times, they may be administered at a set interval, e.g., daily, every other day, weekly, or monthly. Alternatively, they can be administered at an irregular interval, for example on an as-needed basis based on symptoms, patient health, and the like. A therapeutically effective amount of the combination may be administered q.d. for one day, at least 2 days, at least 3 days, at least 4 days, at least 5 days, at least 6 days, at least 7 days, at least 10 days, or at least 15 days. Optionally, the status of the cancer or the regression of the tumor is monitored during or after the treatment, for example, by a FES-PET scan of the subject. The dosage of the combination administered to the subject can be increased or decreased depending on the status of the cancer or the regression of the tumor detected.

[0067] Ideally, the therapeutically effective amount does not exceed the maximum tolerated dosage at which 50% or more of treated subjects experience nausea or other toxicity reactions that prevent further drug administrations. A therapeutically effective amount may vary for a subject depending on a variety of factors, including variety and extent of the

symptoms, sex, age, body weight, or general health of the subject, administration mode and salt or solvate type, variation in susceptibility to the drug, the specific type of the disease, and the like.

[0068] Examples of therapeutically effective amounts of a elacestrant or solvates (e.g., hydrate) or salts thereof for use in the methods disclosed herein include, without limitation, about 150 to about 1,500 mg, about 200 to about 1,500 mg, about 250 to about 1,500 mg, or about 300 to about 1,500 mg dosage q.d. for subjects having resistant ER-driven tumors or cancers; about 150 to about 1,500 mg, about 200 to about 1,000 mg or about 250 to about 1,000 mg or about 300 to about 1,000 mg dosage q.d. for subjects having both wild-type ER driven tumors and/or cancers and resistant tumors and/or cancers; and about 300 to about 500 mg, about 300 to about 550 mg, about 300 to about 600 mg, about 250 to about 500 mg, about 250 to about 550 mg, about 250 to about 600 mg, about 200 to about 500 mg, about 200 to about 550 mg, about 200 to about 600 mg, about 150 to about 500 mg, about 150 to about 550 mg, or about 150 to about 600 mg q.d. dosage for subjects having majorly wild-type ER driven tumors and/or cancers. In certain embodiments, the dosage of a compound of Formula I (e.g., elacestrant) or a salt or solvate thereof for use in the presently disclosed methods general for an adult subject may be approximately 200 mg, 400 mg, 30 mg to 2,000 mg, 100 mg to 1,500 mg, or 150 mg to 1,500 mg p.o., q.d.. This daily dosage may be achieved via a single administration or multiple administrations.

[0069] Dosing of elacestrant in the treatment of breast cancer including resistant strains as well as instances expressing mutant receptor(s) are in the range of 100 mg to 1,000 mg per day. For example, elacestrant may be dosed at 100, 200, 300, 400, 500, 600, 700, 800, 900 or 1,000 mg per day. In particular, 200 mg, 400 mg, 500 mg, 600 mg, 800 mg and 1,000 mg per day are noted. The surprisingly long half-life of elacestrant in humans after PO dosing make this option particularly viable. Accordingly, the drug may be administered as 200 mg bid (400 mg total daily), 250 mg bid (500 mg total daily), 300 mg bid (600 mg total daily), 400 mg bid (800 mg daily) or 500 mg bid (1,000 mg total daily). In some embodiments, the dosing is oral.

[0070] In certain embodiments of the methods disclosed herein, elacestrant or a solvate (e.g., hydrate) or salt thereof preferably accumulate in tumor at a T/P (elacestrant concentration in tumor/elacestrant concentration in plasma) ratio of about 15 or higher, about 18 or higher, about 19 or higher, about 20 or higher, about 25 or higher, about 28 or higher, about 30 or higher, about 33 or higher, about 35 or higher, or about 40 or higher.

[0071] The elacestrant or solvates (e.g., hydrate) or salts thereof may be administered to a subject one time or multiple times. In those embodiments wherein the compounds are administered multiple times, they may be administered at a set interval, e.g., daily, every other day, weekly, or monthly. Alternatively, they can be administered at an irregular interval, for example on an as-needed basis based on symptoms, patient health, and the like.

Formulation

[0072] In some embodiments, elacestrant or solvates (e.g., hydrate) or salts thereof are administered as part of a single formulation. For example, elacestrant or solvates (e.g., hydrate) or salts thereof are formulated in a single pill for oral administration or in a single dose for injection. In certain embodiments, administration of the compounds in a single formulation improves patient compliance.

[0073] In some embodiments, a formulation comprising elacestrant or solvates (e.g., hydrate) or salts thereof may further comprise one or more pharmaceutical excipients, carriers, adjuvants, and/or preservatives.

[0074] The elacestrant or solvates (e.g., hydrate) or salts thereof for use in the presently disclosed methods can be formulated into unit dosage forms, meaning physically discrete units suitable as unitary dosage for subjects undergoing treatment, with each unit containing a predetermined quantity of active material calculated to produce the desired therapeutic effect, optionally in association with a suitable pharmaceutical carrier. The unit dosage form can be for a single daily dose or one of multiple daily doses (e.g., about 1 to 4 or more times q.d.). When multiple daily doses are used, the unit dosage form can be the same or different for each dose. In certain embodiments, the compounds may be formulated for controlled release.

[0075] The elacestrant or solvates (e.g., hydrate) or salts thereof and salts or solvates for use in the presently disclosed methods can be formulated according to any available conventional method. Examples of preferred dosage forms include a tablet, a powder, a subtle granule, a granule, a coated tablet, a capsule, a syrup, a troche, an inhalant, a suppository, an injectable, an ointment, an ophthalmic ointment, an eye drop, a nasal drop, an ear drop, a cataplasm, a lotion and the like. In the formulation, generally used additives such as a diluent, a binder, an disintegrant, a lubricant, a colorant, a flavoring agent, and if necessary, a stabilizer, an emulsifier, an absorption enhancer, a surfactant, a pH adjuster, an antiseptic, an antioxidant and the like can be used. In addition, the formulation is also carried out by combining compositions that are generally used as a raw material for pharmaceutical formulation, according to the conventional methods. Examples of these compositions include, for example, (1) an oil such as a soybean oil, a beef tallow and synthetic glyceride;

(2) hydrocarbon such as liquid paraffin, squalane and solid paraffin; (3) ester oil such as octyldodecyl myristic acid and isopropyl myristic acid; (4) higher alcohol such as cetostearyl alcohol and behenyl alcohol; (5) a silicon resin; (6) a silicon oil; (7) a surfactant such as polyoxyethylene fatty acid ester, sorbitan fatty acid ester, glycerin fatty acid ester, polyoxyethylene sorbitan fatty acid ester, a solid polyoxyethylene castor oil and polyoxyethylene polyoxypropylene block co-polymer; (8) water soluble macromolecule such as hydroxyethyl cellulose, polyacrylic acid, carboxyvinyl polymer, polyethyleneglycol, polyvinylpyrrolidone and methylcellulose; (9) lower alcohol such as ethanol and isopropanol; (10) multivalent alcohol such as glycerin, propyleneglycol, dipropyleneglycol and sorbitol; (11) a sugar such as glucose and cane sugar; (12) an inorganic powder such as anhydrous silicic acid, aluminum magnesium silicicate and aluminum silicate; and (13) purified water, and the like. Additives for use in the above formulations may include, for example, 1) lactose, corn starch, sucrose, glucose, mannitol, sorbitol, crystalline cellulose and silicon dioxide as the diluent; 2) polyvinyl alcohol, polyvinyl ether, methyl cellulose, ethyl cellulose, gum arabic, tragacanth, gelatine, shellac, hydroxypropyl cellulose, hydroxypropylmethyl cellulose, polyvinylpyrrolidone, polypropylene glycol-poly oxyethylene-block co-polymer, meglumine, calcium citrate, dextrin, pectin and the like as the binder; 3) starch, agar, gelatine powder, crystalline cellulose, calcium carbonate, sodium bicarbonate, calcium citrate, dextrin, pectic, carboxymethylcellulose/calcium and the like as the disintegrant; 4) magnesium stearate, talc, polyethyleneglycol, silica, condensed plant oil and the like as the lubricant; 5) any colorants whose addition is pharmaceutically acceptable is adequate as the colorant; 6) cocoa powder, menthol, aromatizer, peppermint oil, cinnamon powder as the flavoring agent; and 7) antioxidants whose addition is pharmaceutically accepted such as ascorbic acid or alpha-tophenol.

[0076] Elacestrant or solvates (e.g., hydrate) or salts thereof for use in the presently disclosed methods can be formulated into a pharmaceutical composition as any one or more of the active compounds described herein and a physiologically acceptable carrier (also referred to as a pharmaceutically acceptable carrier or solution or diluent). Such carriers and solutions include pharmaceutically acceptable salts and solvates of compounds used in the methods of the instant invention, and mixtures comprising two or more of such compounds, pharmaceutically acceptable salts of the compounds and pharmaceutically acceptable solvates of the compounds. Such compositions are prepared in accordance with acceptable pharmaceutical procedures such as described in Remington's Pharmaceutical Sciences, 17th

edition, ed. Alfonso R. Gennaro, Mack Publishing Company, Eaton, Pa. (1985), which is incorporated herein by reference.

[0077] The term "pharmaceutically acceptable carrier" refers to a carrier that does not cause an allergic reaction or other untoward effect in patients to whom it is administered and are compatible with the other ingredients in the formulation. Pharmaceutically acceptable carriers include, for example, pharmaceutical diluents, excipients or carriers suitably selected with respect to the intended form of administration, and consistent with conventional pharmaceutical practices. For example, solid carriers/diluents include, but are not limited to, a gum, a starch (e.g., corn starch, pregelatinized starch), a sugar (e.g., lactose, mannitol, sucrose, dextrose), a cellulosic material (e.g., microcrystalline cellulose), an acrylate (e.g., polymethylacrylate), calcium carbonate, magnesium oxide, talc, or mixtures thereof. Pharmaceutically acceptable carriers may further comprise minor amounts of auxiliary substances such as wetting or emulsifying agents, preservatives or buffers, which enhance the shelf life or effectiveness of the therapeutic agent.

[0078] Elacestrant or solvates (e.g., hydrate) or salts thereof in a free form can be converted into a salt by conventional methods. The term "salt" used herein is not limited as long as the salt is formed with elacestrant or solvates (e.g., hydrate) or salts thereof and is pharmacologically acceptable; preferred examples of salts include a hydrohalide salt (for instance, hydrochloride, hydrobromide, hydroiodide and the like), an inorganic acid salt (for instance, sulfate, nitrate, perchlorate, phosphate, carbonate, bicarbonate and the like), an organic carboxylate salt (for instance, acetate salt, maleate salt, tartrate salt, fumarate salt, citrate salt and the like), an organic sulfonate salt (for instance, methanesulfonate salt, ethanesulfonate salt, benzenesulfonate salt, toluenesulfonate salt, camphorsulfonate salt and the like), an amino acid salt (for instance, aspartate salt, glutamate salt and the like), a quaternary ammonium salt, an alkaline metal salt (for instance, sodium salt, potassium salt and the like), an alkaline earth metal salt (magnesium salt, calcium salt and the like) and the like. In addition, hydrochloride salt, sulfate salt, methanesulfonate salt, acetate salt and the like are preferred as "pharmacologically acceptable salt" of the compounds according to the present invention.

[0079] Isomers of elacestrant or solvates (e.g., hydrate) or salts thereof (e.g., geometric isomers, optical isomers, rotamers, tautomers, and the like) can be purified using general separation means, including for example recrystallization, optical resolution such as diastereomeric salt method, enzyme fractionation method, various chromatographies (for instance, thin layer chromatography, column chromatography, glass chromatography and the

like) into a single isomer. The term "a single isomer" herein includes not only an isomer having a purity of 100%, but also an isomer containing an isomer other than the target, which exists even through the conventional purification operation. A crystal polymorph sometimes exists for elacestrant or solvates (e.g., hydrate) or salts thereof and/or fulvestrant, and all crystal polymorphs thereof are included in the present invention. The crystal polymorph is sometimes single and sometimes a mixture, and both are included herein.

[0080] In certain embodiments, elacestrant or solvates (e.g., hydrate) or salts thereof may be in a prodrug form, meaning that it must undergo some alteration (e.g., oxidation or hydrolysis) to achieve its active form. Alternative, elacestrant or solvates (e.g., hydrate) or salts thereof may be a compound generated by alteration of a parental prodrug to its active form.

Administration Route

[0081] Administration routes of elacestrant or solvates (e.g., hydrate) or salts thereof include but not limited to topical administration, oral administration, intradermal administration, intramuscular administration, intraperitoneal administration, intravenous administration, intravesical infusion, subcutaneous administration, transdermal administration, and transmucosal administration. In some embodiments, the administration route is oral.

Gene Profiling

[0082] In certain embodiments, the methods of tumor growth inhibition or tumor regression provided herein further comprise gene profiling the subject, wherein the gene to be profiled is one or more genes selected from the group consisting of ABL1, AKT1, AKT2, ALK, APC, AR, ARID1A, ASXL1, ATM, AURKA, BAP, BAP1, BCL2L11, BCR, BRAF, BRCA1, BRCA2, CCND1, CCND2, CCND3, CCNE1, CDH1, CDK4, CDK6, CDK8, CDKN1A, CDKN1B, CDKN2A, CDKN2B, CEBPA, CTNNB1, DDR2, DNMT3A, E2F3, EGFR, EML4, EPHB2, ERBB2, ERBB3, ESR1, EWSR1, FBXW7, FGF4, FGFR1, FGFR2, FGFR3, FLT3, FRS2, HIF1A, HRAS, IDH1, IDH2, IGF1R, JAK2, KDM6A, KDR, KIF5B, KIT, KRAS, LRP1B, MAP2K1, MAP2K4, MCL1, MDM2, MDM4, MET, MGMT, MLL, MPL, MSH6, MTOR, MYC, NF1, NF2, NKX2-1, NOTCH1, NPM, NRAS, PDGFRA, PIK3CA, PIK3R1, PML, PTEN, PTPRD, RARA, RB1, RET, RICTOR, ROS1, RPTOR, RUNX1, SMAD4, SMARCA4, SOX2, STK11, TET2, TP53, TSC1, TSC2, and VHL. In other embodiments, the gene to be profiled is one or more genes selected from the group consisting of AKT1, AKT2, BRAF, CDK4, CDK6, PIK3CA, PIK3R1, and MTOR.

[0083] In some embodiments, this invention provides a method of treating a subpopulation of breast cancer patients wherein said sub-population has increased expression of one or more of the genes disclosed supra, and treating said sub-population with an effective dose of elacestrant or solvates (e.g., hydrate) or salts thereof according to the dosing embodiments as described in this disclosure.

Dose Adjusting

[0084] In addition to establishing the ability of elacestrant to inhibit tumor growth, elacestrant inhibits estradiol binding to ER in the uterus and pituitary. In these experiments, estradiol binding to ER in uterine and pituitary tissue was evaluated by FES-PET imaging. After treatment with elacestrant, the observed level of ER binding was at or below background levels. These results establish that the antagonistic effect of elacestrant on ER activity can be evaluated using real-time scanning. Based on these results, methods are provided herein for monitoring the efficacy of treatment elacestrant or solvates (e.g., hydrate) or salts thereof in a combination therapy disclosed herein by measuring estradiol-ER binding in one or more target tissues, wherein a decrease or disappearance in binding indicates efficacy.

[0085] Further provided are methods of adjusting the dosage of elacestrant or solvates (e.g., hydrate) or salts thereof in a combination therapy disclosed herein based on estradiol-ER binding. In certain embodiments of these methods, binding is measured at some point following one or more administrations of a first dosage of the compound. If estradiol-ER binding is not affected or exhibits a decrease below a predetermined threshold (e.g., a decrease in binding versus baseline of less than 5%, less than 10%, less than 20%, less than 30%, or less than 50%), the first dosage is deemed to be too low. In certain embodiments, these methods comprise an additional step of administering an increased second dosage of the compound. These steps can be repeated, with dosage repeatedly increased until the desired reduction in estradiol-ER binding is achieved. In certain embodiments, these steps can be incorporated into the methods of inhibiting tumor growth provided herein. In these methods, estradiol-ER binding can serve as a proxy for tumor growth inhibition, or a supplemental means of evaluating growth inhibition. In other embodiments, these methods can be used in conjunction with the administration of elacestrant or solvates (e.g., hydrate) or salts thereof for purposes other than inhibition of tumor growth, including for example inhibition of cancer cell proliferation.

[0086] In certain embodiments, the methods provided herein for adjusting the dosage of elacestrant or salt or solvate (e.g., hydrate) thereof in a combination therapy comprise:

(1) administering a first dosage of elacestrant or salt or solvate (e.g., hydrate) thereof (e.g., about 350 to about 500 or about 200 to about 600 mg/day) for 3, 4, 5, 6, or 7 days;

(2) detecting estradiol-ER binding activity; wherein:

(i) if the ER binding activity is not detectable or is below a predetermined threshold level, continuing to administer the first dosage (i.e., maintain the dosage level); or

(ii) if the ER binding activity is detectable or is above a predetermined threshold level, administering a second dosage that is greater than the first dosage (e.g., the first dosage plus about 50 to about 200 mg) for 3, 4, 5, 6, or 7 days, then proceeding to step (3);

(3) detecting estradiol-ER binding activity; wherein

(i) if the ER binding activity is not detectable or is below a predetermined threshold level, continuing to administer the second dosage (i.e., maintain the dosage level); or

(ii) if the ER binding activity is detectable or is above a predetermined threshold level, administering a third dosage that is greater than the second dosage (e.g., the second dosage plus about 50 to about 200 mg) for 3, 4, 5, 6, or 7 days, then proceeding to step (4);

(4) repeating the steps above through a fourth dosage, fifth dosage, etc., until no ER binding activity is detected.

[0087] In certain embodiments, the invention includes the use of PET imaging to detect and/or dose ER sensitive or ER resistant cancers.

[0088] The following examples are provided to better illustrate the claimed invention and are not to be interpreted as limiting the scope of the invention. To the extent that specific materials are mentioned, it is merely for purposes of illustration and is not intended to limit the invention. One skilled in the art may develop equivalent means or reactants without the exercise of inventive capacity and without departing from the scope of the invention. It will be understood that many variations can be made in the procedures herein described while still remaining within the bounds of the present invention. It is the intention of the inventors that such variations are included within the scope of the invention.

Examples

Materials and Methods

Test compounds

[0089] Elacestrant used in the examples below was (6R)-6-(2-(N-(4-(2-(ethylamino)ethyl)benzyl)-N-ethylamino)-4-methoxyphenyl)-5,6,7,8-tetrahydronaphthalen-2-ol dihydrochloride, manufactured by, for example, IRIX Pharmaceuticals, Inc. (Florence, SC). Elacestrant was stored as a dry powder, formulated for use as a homogenous suspension in 0.5% (w/v) methylcellulose in deionized water, and for animal models was administered p.o.. Tamoxifen, raloxifene and estradiol (E2) were obtained from Sigma-Aldrich (St. Louis, MO), and administered by subcutaneous injection. Fulvestrant was obtained from Tocris Biosciences (Minneapolis, MN) and administered by subcutaneous injection. Other laboratory reagents were purchased from Sigma-Aldrich unless otherwise noted.

PDX Models

[0090] Tumors were passaged as fragments into athymic nude mice (Nu (NCR)-Foxn1nu). CTG-1211 (Champions Oncology), ST2535 (START), and WHIM43 (Horizon) patient-derived xenograft fragments were implanted into mice without estradiol supplementation. All mice were housed in pathogen-free housing in individually ventilated cages with sterilized and dust-free bedding cobs, access to sterilized food and water ad libitum, under a light dark cycle (12-14 hour circadian cycle of artificial light) and controlled room temperature and humidity. Tumors were measured twice/wk with Vernier calipers; volumes were calculated using the formula: (L*W2)*0.52.Elacestrant was administered orally, daily for duration of study. Fulvestrant was administered once/week subcutaneously.

Quantitative Real-time PCR (RT-qPCR)

***In vivo* Xenograft Models**

[0091] End of study tumors were pulverized with the cryoPREP™ Impactor (Covaris) and total RNA was extracted with the RNeasy mini kit (Qiagen). qPCR was performed using the Taqman Fast Virus 1-Step Master Mix and TaqMan™ probes (Applied Biosystems) The Ct values were analyzed to assess relative changes in expression of PgR (progesterone receptor) mRNA, with GAPDH as an internal control, using the 2-ΔΔCT method.

***In vitro* Xenograft Models**

[0092] At the end of treatment, cells were lysed with the lysis buffer from the 1-step Cells-to-Ct kit and the lysates were processed according to the manufacturer's instructions. qPCR was performed using the 1-step master mix and TaqMan™ probes (Applied Biosystems).

The Ct values were analyzed to assess relative changes in expression of PgR (progesterone receptor) mRNA, with GAPDH as an internal control, using the 2-ΔΔCT method.

Agent Efficacy

[0093] For all studies, beginning Day 0, tumor dimensions were measured by digital caliper and data including individual and mean estimated tumor volumes (Mean TV ± SEM) recorded for each group; tumor volume was calculated using the formula (Yasui et al. Invasion Metastasis 17:259-269 (1997), which is incorporated herein by reference): *TV= width² x length x 0.52.* Each group or study was ended once the estimated group mean tumor volume reached the Tumor Volume (TV) endpoint (time endpoint was 60 days; and volume endpoint was group mean 2 cm^3); individual mice reaching a tumor volume of 2 cm^3 or more were removed from the study and the final measurement included in the group mean until the mean reached volume endpoint or the study reached time endpoint.

Efficacy Calculations and Statistical Analysis

[0094] %Tumor Growth Inhibition (%TGI) values were calculated at a single time point (when the control group reached tumor volume or time endpoint) and reported for each treatment group (T) versus control (C) using initial (i) and final (f) tumor measurements by the formula (Corbett TH et al. *In vivo* methods for screening and preclinical testing. In: Teicher B, ed., *Anticancer Drug Development Guide.* Totowa, NJ: Humana. 2004: 99-123.): *%TGI= 1- Tf-Ti / Cf-Ci.*

Statistical Analysis

[0095] Statistical analysis was performed using GraphPadPrism 7.0 and data is generally expressed as mean ± SEM/SD. Treatment group comparisons were performed using one way ANOVA statistical analyses was performed with a Dunnett's post-test. Statistics are expressed as: ns, not significant; *p < 0.05; **p < 0.01; ***p < 0.001; ****p<0.0001).

Sample Collection

[0096] At endpoint, tumors were removed. One fragment was flash frozen, while another fragment was placed in 10% NBF for at least 24 hours and formalin fixed paraffin embedded (FFPE). Flash frozen samples were stored at -80°C; FFPE blocks were stored at room temperature.

Western Blot

[0097] Cells or tumors were harvested post-dosing and protein expression analyzed using standard practice and antibodies as follows: ERa, PR, (Cell Signaling Technologies, Cat#13258; #3153) and Vinculin: Sigma-Aldrich, #v9131). Protein expression was quantified using the AzureSpot software and normalized to vinculin expression.

Examples

[0098] Referring to FIG. 1, elacestrant was demonstrated to inhibit proliferation and ER signaling in *in vitro* models harboring various ESR1 mutations, including Y537S clone 1, Y537S clone 2, D538G clone 1, D538G clone 2, and S463P clone 1 cancer cell lines. The representative pictures presented in the top row visualize tumor cells treated with vehicle control for the Y537S clone 1, Y537S clone 2, D538G clone 1, D538G clone 2, and S463P clone 1 mutated cancer cell lines. The pictures presented in the bottom row visualize the Y537S clone 1, Y537S clone 2, D538G clone 1, D538G clone 2, and S463P clone 1 tumor cells treated with elacestrant at 100 nM.

[0099] Referring now to FIG. 2, elacestrant demonstrated dose-dependent inhibition of tumor growth and tumor regression in athymic nude mice xenograft models. In FIG. 2, mean +/- SEM tumor volumes over time in mouse xenograft models were treated with vehicle control, elacestrant (30, 60, and 120 mg/kg) and fulvestrant (1 mg/dose).

[00100] Referring now to FIGS. 3A-3C, elacestrant was demonstrated to inhibit ER signaling in *in vitro* models harboring various ESR1 mutations where the representative histograms show a decrease of proliferation markers in xenograft models *in vitro*. In FIG. 3A, fold change relative to control of progesterone receptor (PgR) is provided for tumor cell models having wild type, S463P, D538G, and Y537S mutations treated with vehicle control, elacestrant (10, 100, and 1000 nM), E2 (10pM), and fulvestrant (10, 100, 1000 nM). In FIG. 3B, fold change relative to control of growth regulated by estrogen (GREB1) is provided in tumor cell models having wild type, S463P, D538G, and Y537S mutations treated with vehicle control, elacestrant (10, 100, and 1000 nM), E2 (10pM), and fulvestrant (10, 100, 1000 nM). In FIG. 3C, fold change relative to control of trefoil factor 1 (TFF1) is provided in tumor cell models having wild type, S463P, D538G, and Y537S mutations treated with vehicle control, elacestrant (10, 100, and 1000 nM), E2 (10pM), and fulvestrant (10, 100, 1000 nM).

[00101] Referring now to FIGS. 4A-4C, elacestrant demonstrated dose-dependent inhibition of tumor growth in multiple PDX models harboring the ESR1:D538G mutation. In FIG. 4A, mean +/- SEM tumor volumes over time in athymic nude mice implanted with the ST2535-HI PDX xenograft (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation were treated with vehicle control and elacestrant (30 and 60 mg/kg). In FIG. 4B, mean +/- SEM tumor volumes over time in athymic nude mice implanted with the CTG-1211-HI PDX xenograft (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation were treated with

vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose). In FIG. 4C, mean +/- SEM tumor volumes over time in athymic nude mice implanted with the WHIM43-HI PDX xenograft (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation were treated with vehicle control, elacestrant (30 and 60 mg/kg) and fulvestrant (3 mg/dose).

[00102] Referring now to FIGS. 5A-5F, elacestrant was demonstrated to degrade ER and inhibit ER signaling in PDX models harboring ESR1:D538G mutations in athymic nude mice xenograft models. In FIG. 5A, fold change over vehicle control of progesterone receptor (PgR) mRNA levels in the ST2535-HI PDX xenograft model (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation was treated with vehicle control and elacestrant (30 and 60 mg/kg). In FIG. 5B, a Western blot is illustrated showing PgR expression in the ST2535-HI PDX xenograft model with an ESR1:D538G mutation treated with vehicle control and elacestrant (30 and 60 mg/kg). In FIG. 5C, fold change over vehicle control of progesterone receptor (PgR) mRNA levels in the CTG-1211-HI PDX xenograft model (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation was treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose). In FIG. 5D, a Western blot is illustrated showing PgR expression in the CTG-1211-HI PDX xenograft model with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant. In FIG. 5E, fold change over vehicle control of progesterone receptor (PgR) mRNA levels in the WHIM43-HI PDX xenograft model (previously treated with tamoxifen, aromatase inhibitor, and fulvestrant) with an ESR1:D538G mutation was treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose). In FIG. 5F, a Western blot is illustrated showing PgR expression in the WHIM43-HI PDX xenograft model with an ESR1:D538G mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant.

[00103] Referring now to FIGS. 6A-6B, elacestrant demonstrated greater tumor growth inhibition than comparator SERDs in ST941-HI PDX models harboring ESR1:Y537S mutations. In FIG. 6A, mean +/- SEM tumor volumes over time in the ST941-HI PDX model harboring an ESR1:Y537S mutation was treated with vehicle control, elacestrant (10, 30, and 60 mg/kg) fulvestrant (3 mg/dose, s.c., q.d.), serd1 dose 1, and serd1 dose 2. In FIG. 6B, mean +/- SEM tumor volumes over time in the ST941-HI PDX model harboring an ESR1:Y537S mutation was treated with vehicle control, elacestrant (10, 30, and 60 mg/kg) fulvestrant (3 mg/dose), serd2 dose 1, and serd2 dose 2.

[00104] Referring now to FIGS. 7A-7C, elacestrant demonstrated greater tumor growth inhibition than comparator SERDs in ST941-HI PDX models harboring ESR1:Y537S mutations. In FIG. 7A, fold change over vehicle control relative to progesterone receptor (PgR) mRNA levels in the ST941-HI PDX model harboring an ESR1:Y537S mutation treated with vehicle control, fulvestrant (3 mg/dose), elacestrant (30 mg/kg), serd1 dose 1, serd1 dose 2, serd2 dose 1, and serd2 dose 2. In FIG. 7B, a Western blot is provided for the ST941-HI PDX model harboring an ESR1:Y537S mutation demonstrating PgR expression that was treated with vehicle control, fulvestrant (3 mg/kg), elacestrant (30 mg/kg), serd1 dose 1, and serd1 dose 2. In FIG. 7C, a Western blot is provided for the ST941-HI PDX model harboring an ESR1:Y537S mutation demonstrating PgR expression that was treated with vehicle control, fulvestrant (3 mg/kg), elacestrant (30 mg/kg), serd2 dose 1, and serd2 dose 2.

[00105] Referring to FIGS. 8A-8B, the evaluation of elacestrant and fulvestrant and their *in vitro* respective activities are provided. In FIG. 8A, *in vitro* cell viability (% of control) is provided with respect to Log[Concentration (μm)] for a ST941-HI PDX cell line. In FIG. 8B, fold change over vehicle control of progesterone receptor (PgR) mRNA levels is plotted with respect to the concentration of elacestrant (0, 10, 100, and 1000 nM) and fulvestrant (0, 10, 100, and 1000 nM) used in treating *in vitro* ST941-HI cell line derived from PDX.

[00106] Referring to FIGS. 9A-9B, the evaluation of elacestrant and fulvestrant and their *in vivo* respective activities are provided. In FIG. 9A, mean +/- SEM tumor volumes in mice implanted with the ST941-HI PDX harboring an ESR1:Y537S mutation are plotted with respect to time and their treatment with vehicle control, elacestrant (10, 30, and 60 mg/kg) and fulvestrant (3 mg/dose). In FIG. 9B, fold change relative to control of progesterone receptor (PgR) mRNA expression in the ST941-HI PDX model are plotted with respect to their treatment with vehicle control, fulvestrant (3 mg/dose), and elacestrant (10, 30, and 60 mg/kg).

[00107] Referring now to FIGS. 10A-10D, elacestrant and fulvestrant demonstrate partial efficacy in an ESR1 mutant PDX model harboring additional oncogenic mutations. In FIG. 10A, mean +/- SEM tumor volumes over time in mice implanted with the WHIM20 PDX xenograft with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30, and 60 mg/kg) and fulvestrant (3 mg/dose). In FIG. 10B, fold change relative to vehicle control of progesterone receptor (PgR) in the WHIM20 PDX xenograft with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose) is provided. In FIG. 10C, fold change relative to control of trefoil factor 1 (TFF1)

in the WHIM20 PDX xenograft with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose) is provided. In FIG. 10D, fold change relative to control of growth regulated by estrogen (GREB1) in the WHIM20 PDX xenograft with an ESR1:Y537S^{hom} mutation treated with vehicle control, elacestrant (30 and 60 mg/kg), and fulvestrant (3 mg/dose) is provided. In FIG. 10E, a Western blot of a WHIM20 PDX xenograft with an ESR1:Y537S^{hom} mutation showing PgR expression and treated with vehicle control, fulvestrant (3 mg/dose), and elacestrant (30 and 60 mg/kg) is provided.

OTHER EMBODIMENTS

[00108] All publications and patents referred to in this disclosure are incorporated herein by reference to the same extent as if each individual publication or patent application were specifically and individually indicated to be incorporated by reference. Should the meaning of the terms in any of the patents or publications incorporated by reference conflict with the meaning of the terms used in this disclosure, the meaning of the terms in this disclosure are intended to be controlling. Furthermore, the foregoing discussion discloses and describes merely exemplary embodiments of the present invention. One skilled in the art will readily recognize from such discussion and from the accompanying drawings and claims, that various changes, modifications and variations can be made therein without departing from the spirit and scope of the invention as defined in the following claims.

WHAT IS CLAIMED IS:

1. A method of inhibiting and degrading a mutant estrogen receptor alpha positive cancer in a subject comprising administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof.

2. The method of claim 1, wherein the mutant estrogen receptor alpha positive cancer comprises one or more mutations selected from the group consisting of D538G, $Y537X_1$, $L536X_2$, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: X_1 is S, N, or C; and X_2 is R or Q.

3. The method of claim 2, wherein the mutation is Y537S.

4. The method of claim 2, wherein the mutation is D538G.

5. The method of any one of claims 1-4, wherein the mutant estrogen receptor alpha positive cancer is resistant to a drug selected from the group consisting of anti-estrogens, aromatase inhibitors, and combinations thereof.

6. The method of any one of claims 1-5, wherein the mutant estrogen receptor alpha positive cancer is selected from the group consisting of breast cancer, uterine cancer, ovarian cancer, and pituitary cancer.

7. The method of any one of claims 1-6, wherein the mutant estrogen receptor alpha positive cancer is advanced or metastatic breast cancer.

8. The method of any one of claims 1-7, wherein the mutant estrogen receptor alpha positive cancer is breast cancer.

9. The method of any one of claims 1-8, wherein the subject is a post-menopausal woman.

10. The method of any one of claims 1-8, wherein the subject is a pre-menopausal woman.

11. The method of any one of claims 1-8, wherein the subject is a post-menopausal woman who had relapsed or progressed after previous treatment with selective estrogen receptor modulators (SERMs) and/or aromatase inhibitors (AIs).

12. The method of any one of claims 1-11, wherein the elacestrant is administered to the subject at a dose of from about 200 mg/day to about 500 mg/day.

13. The method of any one of claims 1-12, wherein the elacestrant is administered to the subject at a dose of about 200 mg/day, about 300 mg/day, about 400 mg/day, or about 500 mg/day.

14. The method of any one of claims 1-11, wherein the elacestrant is administered to the subject at a dose that is the maximum tolerated dose for the subject.

15. The method of any one of claims 1-14, the method further comprising:

identifying the subject for treatment by measuring increased expression of one or more genes selected from ABL1, AKT1, AKT2, ALK, APC, AR, ARID1A, ASXL1, ATM, AURKA, BAP, BAP1, BCL2L11, BCR, BRAF, BRCA1, BRCA2, CCND1, CCND2, CCND3, CCNE1, CDH1, CDK4, CDK6, CDK8, CDKN1A, CDKN1B, CDKN2A, CDKN2B, CEBPA, CTNNB1, DDR2, DNMT3A, E2F3, EGFR, EML4, EPHB2, ERBB2, ERBB3, ESR1, EWSR1, FBXW7, FGF4, FGFR1, FGFR2, FGFR3, FLT3, FRS2, HIF1A, HRAS, IDH1, IDH2, IGF1R, JAK2, KDM6A, KDR, KIF5B, KIT, KRAS, LRP1B, MAP2K1, MAP2K4, MCL1, MDM2, MDM4, MET, MGMT, MLL, MPL, MSH6, MTOR, MYC, NF1, NF2, NKX2-1, NOTCH1, NPM, NRAS, PDGFRA, PIK3CA, PIK3R1, PML, PTEN, PTPRD, RARA, RB1, RET, RICTOR, ROS1, RPTOR, RUNX1, SMAD4, SMARCA4, SOX2, STK11, TET2, TP53, TSC1, TSC2, and VHL.

16. The method according to claim 15, wherein the one or more genes is selected from AKT1, AKT2, BRAF, CDK4, CDK6, PIK3CA, PIK3R1, and MTOR.

17. The method of any one of claims 1-16, wherein the ratio of the concentration of elacestrant or a salt or solvate thereof in the tumor to the concentration of elacestrant or a salt or solvate thereof in plasma (T/P) following administration is at least about 15.

18. A method of treating a drug resistant estrogen receptor alpha-positive cancer in a subject having a mutant estrogen receptor alpha, the method comprising:

administering to the subject a therapeutically effective amount of elacestrant, or a pharmaceutically acceptable salt or solvate thereof,

wherein the mutant estrogen receptor alpha comprises one or more mutations selected from the group consisting of D538G, Y537X_1, L536X_2, P535H, V534E, S463P, V392I, E380Q and combinations thereof, wherein: X_1 is S, N, or C; and X_2 is R or Q.

19. The method of claim 18, wherein the cancer is resistant to a drug selected from the group consisting of anti-estrogens, aromatase inhibitors, and combinations thereof.

20. The method of claim 19, wherein the anti-estrogens are selected from the group consisting of tamoxifen, toremifene and fulvestrant and the aromatase inhibitors are selected from the group consisting of exemestane, letrozole and anastrozole.

21. The method of any one of claims 18-20, wherein the drug resistant estrogen receptor alpha-positive cancer is selected from the group consisting of breast cancer, uterine cancer, ovarian cancer, and pituitary cancer.

22. The method of any one of claims 18-21, wherein the cancer is advanced or metastatic breast cancer.

23. The method of any one of claims 18-21, wherein the cancer is breast cancer.

24. The method of any one of claims 18-23, wherein the subject is a post-menopausal woman.

25. The method of any one of claims 18-23, wherein the subject is a pre-menopausal woman.

26. The method of any one of claims 18-23, wherein the subject is a post-menopausal woman who had relapsed or progressed after previous treatment with SERMs and/or AIs.

27. The method of any one of claims 18-26, wherein said subject expresses at least one mutant estrogen receptor alpha selected from the group consisting of D538G, Y537S, Y537N, Y537C, E380Q, S463P, L536R, L536Q, P535H, V392I and V534E.

28. The method of any one of claims 18-27, wherein the mutation includes Y537S.

29. The method of any one of claims 18-28, wherein the mutation includes D538G.

30. The method of any one of claims 18-29, the method further comprising:

identifying the subject for treatment by measuring increased expression of one or more genes selected from ABL1, AKT1, AKT2, ALK, APC, AR, ARID1A, ASXL1, ATM, AURKA, BAP, BAP1, BCL2L11, BCR, BRAF, BRCA1, BRCA2, CCND1, CCND2, CCND3, CCNE1, CDH1, CDK4, CDK6, CDK8, CDKN1A, CDKN1B, CDKN2A, CDKN2B, CEBPA, CTNNB1, DDR2, DNMT3A, E2F3, EGFR, EML4, EPHB2, ERBB2, ERBB3, ESR1, EWSR1, FBXW7, FGF4, FGFR1, FGFR2, FGFR3, FLT3, FRS2, HIF1A, HRAS, IDH1, IDH2, IGF1R, JAK2, KDM6A, KDR, KIF5B, KIT, KRAS, LRP1B, MAP2K1, MAP2K4, MCL1, MDM2, MDM4, MET, MGMT, MLL, MPL, MSH6, MTOR, MYC, NF1, NF2, NKX2-1, NOTCH1, NPM, NRAS, PDGFRA, PIK3CA, PIK3R1, PML,

PTEN, PTPRD, RARA, RB1, RET, RICTOR, ROS1, RPTOR, RUNX1, SMAD4, SMARCA4, SOX2, STK11, TET2, TP53, TSC1, TSC2, and VHL.

31. The method according to claim 30, wherein the one or more genes is selected from AKT1, AKT2, BRAF, CDK4, CDK6, PIK3CA, PIK3R1, and MTOR.

32. The method of any one of claims 18-31, wherein the elacestrant is administered to the subject at a dose of from about 200 to about 500 mg/day.

33. The method of any one of claims 18-32, wherein the elacestrant is administered to the subject at a dose of about 200 mg, about 300 mg, about 400 mg, or about 500 mg.

34. The method of any one of claims 18-33, wherein the elacestrant is administered to the subject at a dose of about 300 mg/day.

35. The method of any one of claims 18-34, wherein the ratio of the concentration of elacestrant or a salt or solvate thereof in the tumor to the concentration of elacestrant or a salt or solvate thereof in plasma (T/P) following administration is at least about 15.

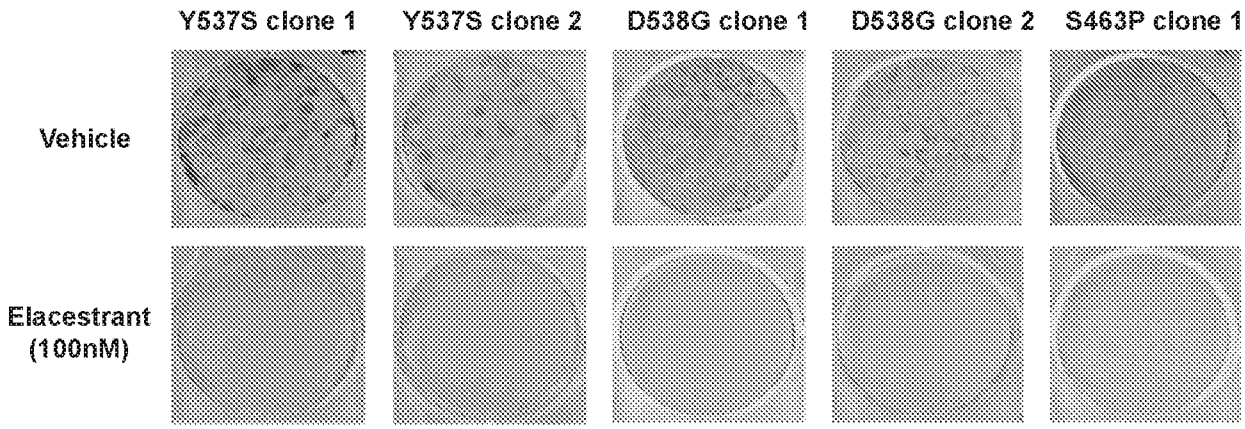

Y537S clone 1
Y537S clone 2
D538G clone 1
D538G clone 2
S463P clone 1
Vehicle
Elacestrant (100nM)

FIG. 1

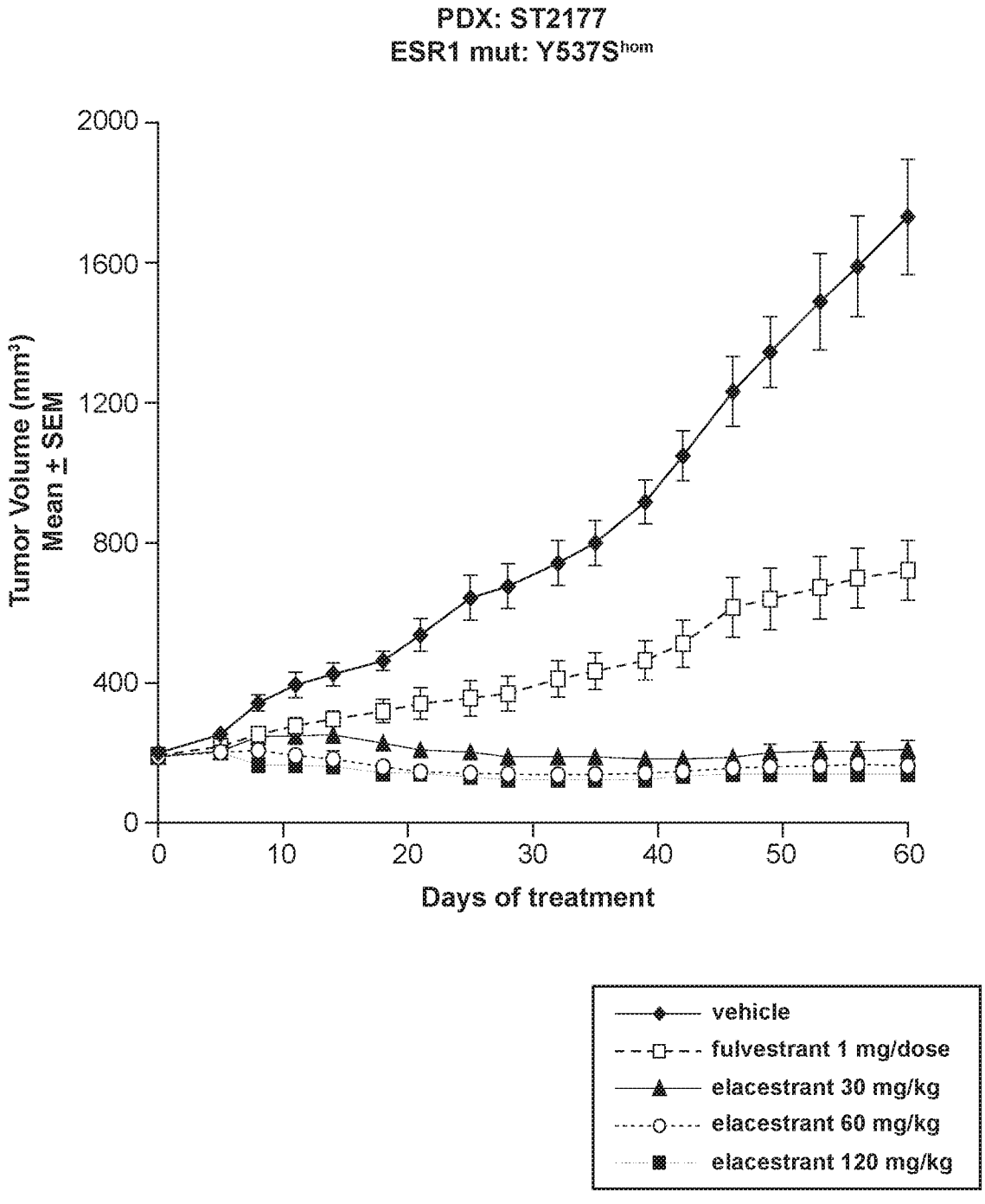

PDX: ST2177
ESR1 mut: Y537S^hom
Tumor Volume (mm³)
Mean ± SEM
2000
1600
1200
800
400
0
0
10
20
30
40
50
60
Days of treatment
vehicle
fulvestrant 1 mg/dose
elacestrant 30 mg/kg
elacestrant 60 mg/kg
elacestrant 120 mg/kg

FIG. 2

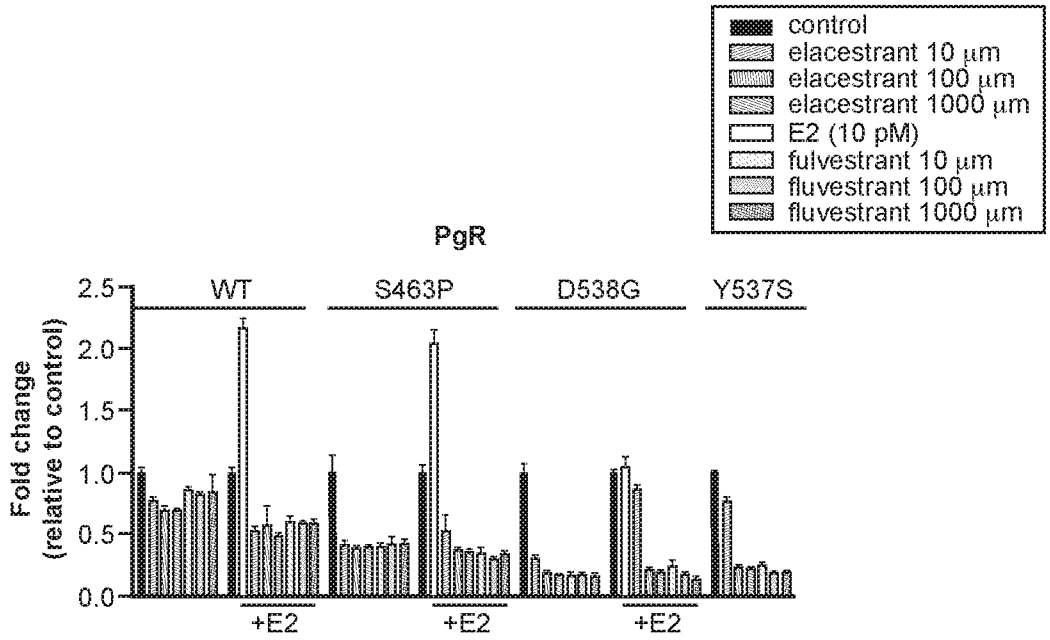


FIG. 3A

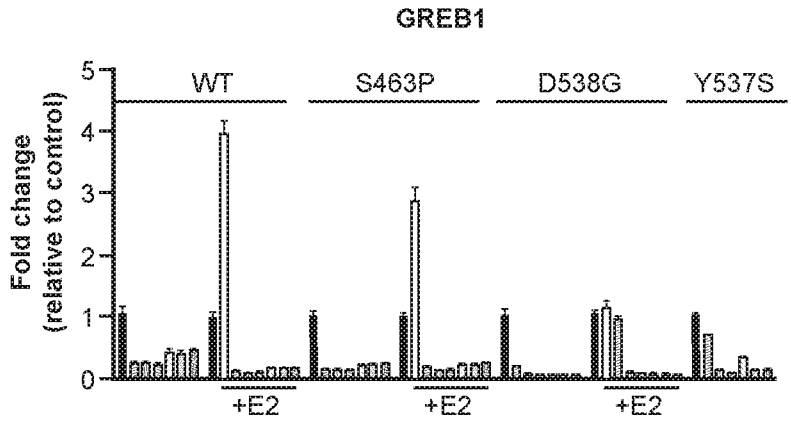


FIG. 3B

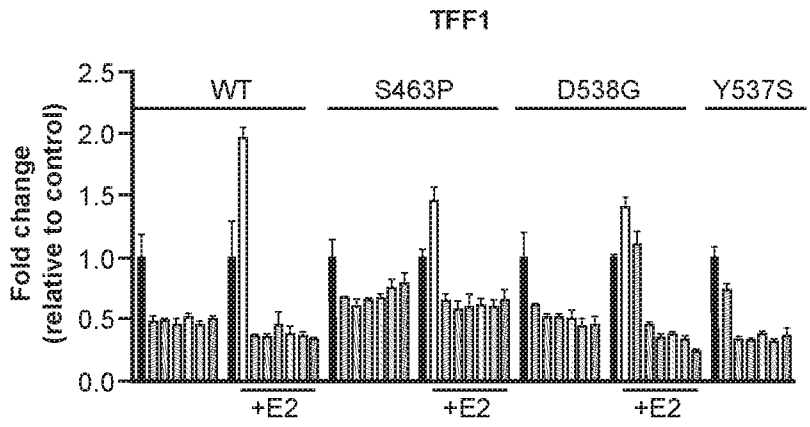
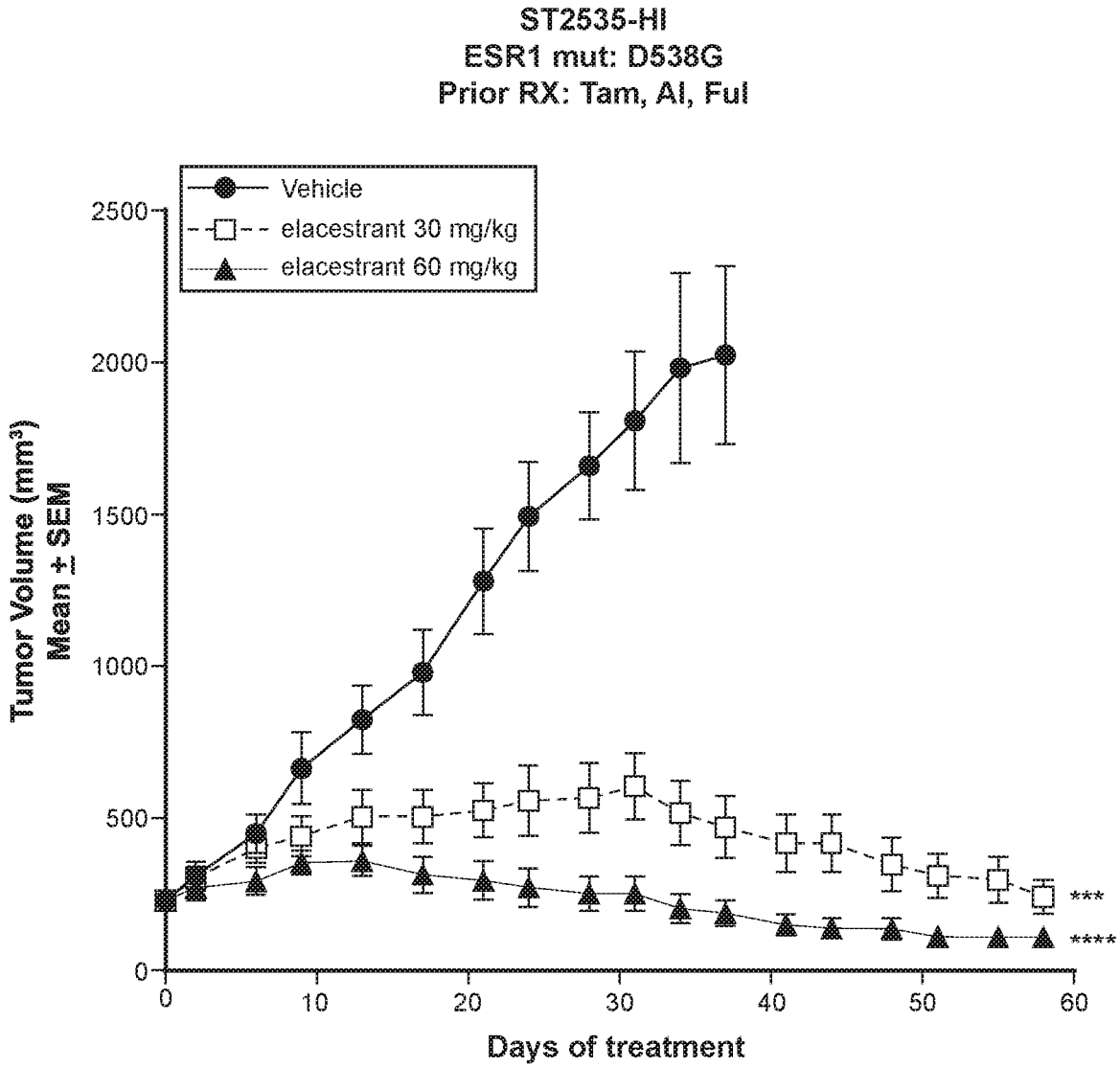
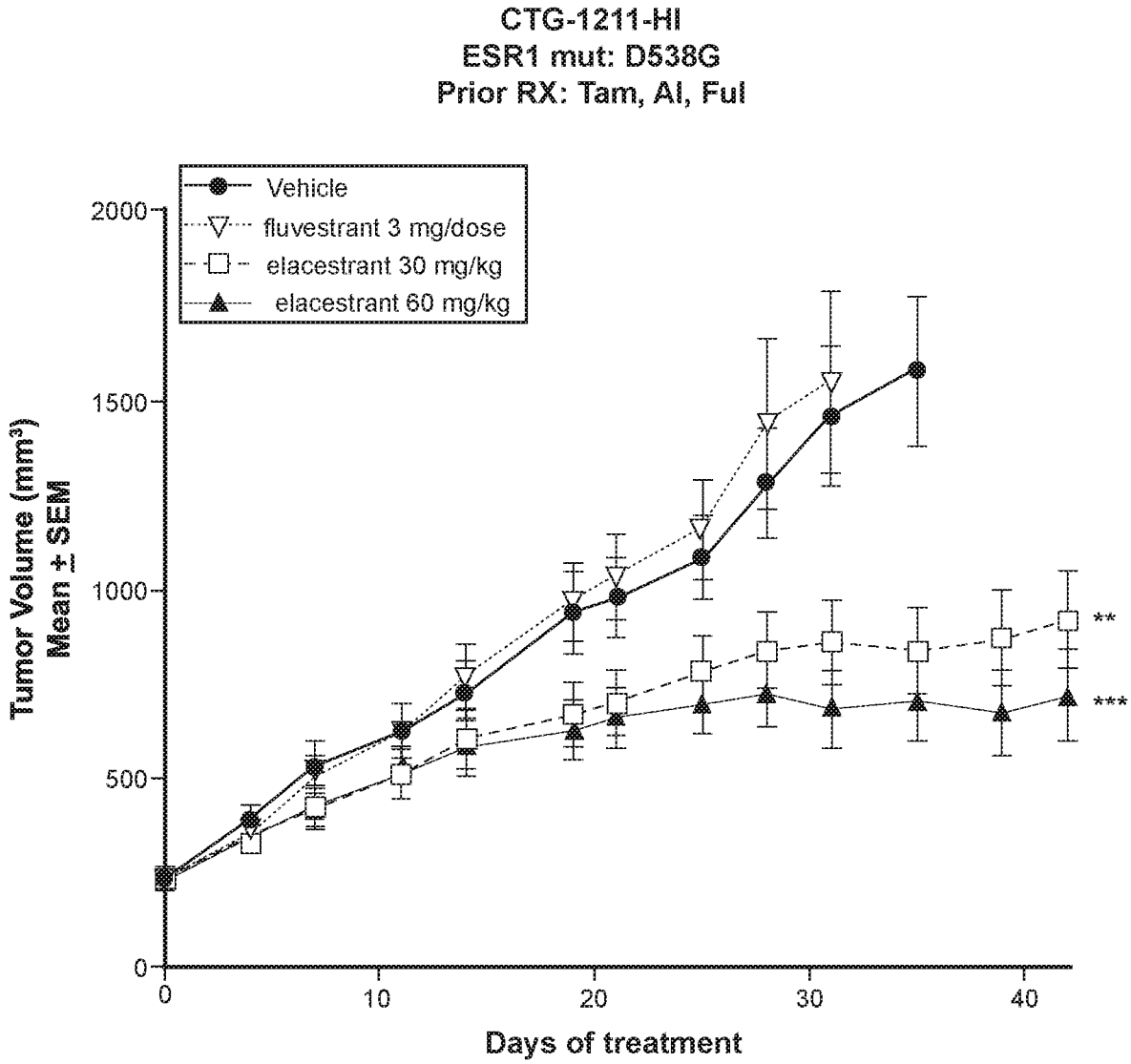


FIG. 3C



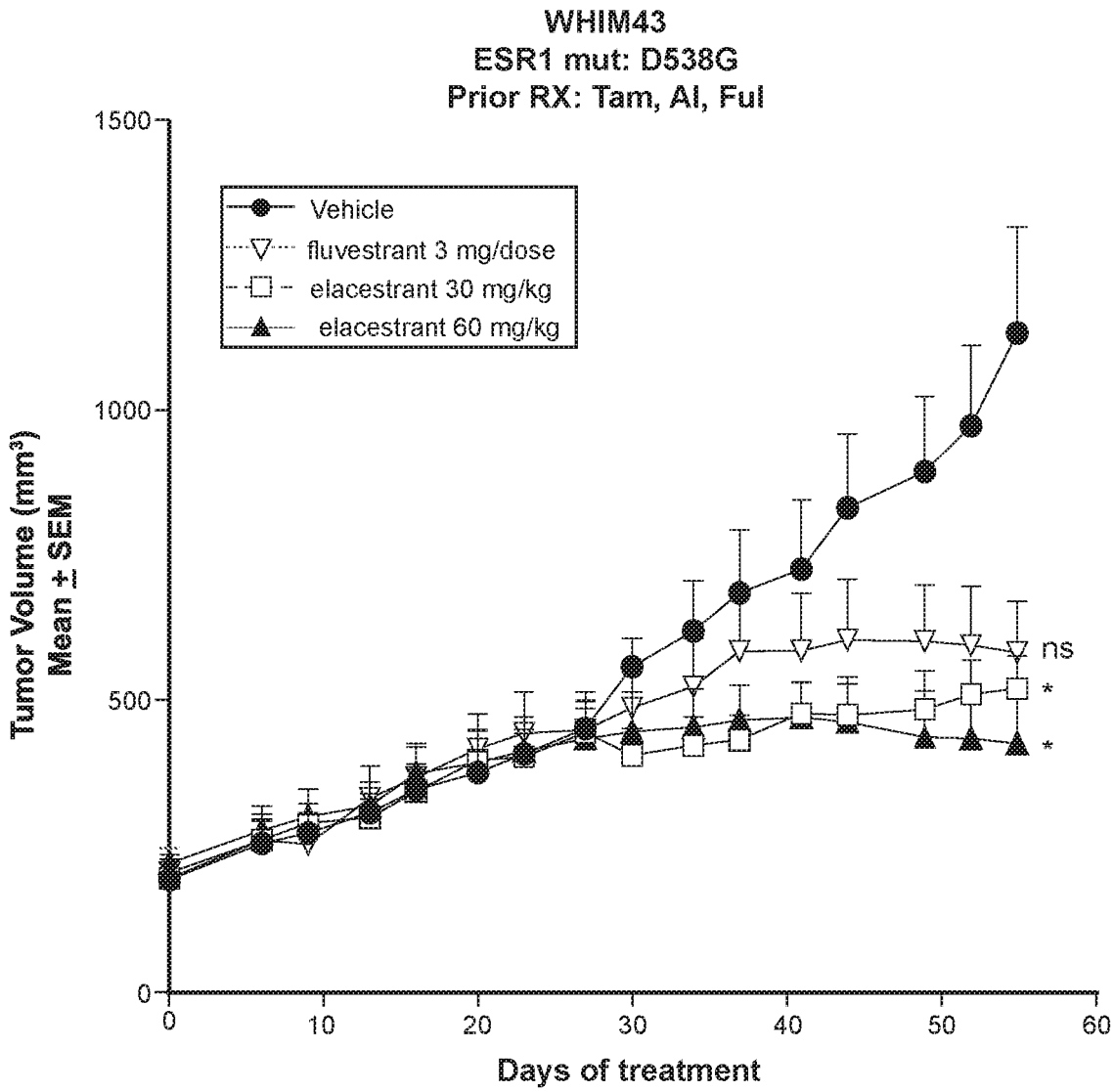
ns = not significant, *p < 0.05, **p < 0.01, ***p < 0.001, ****p < 0.0001
mut = mutant; Tam = tamoxifen; AI = aromatase inhibitor; Ful = fulvestrant

FIG. 4A



ns = not significant, *p < 0.05, **p < 0.01, ***p < 0.001, ****p < 0.0001
mut = mutant; Tam = tamoxifen; AI = aromatase inhibitor; Ful = fulvestrant

FIG. 4B



ns = not significant, *p < 0.05, **p < 0.01, ***p < 0.001, ****p < 0.0001
mut = mutant; Tam = tamoxifen; AI = aromatase inhibitor; Ful = fulvestrant

FIG. 4C

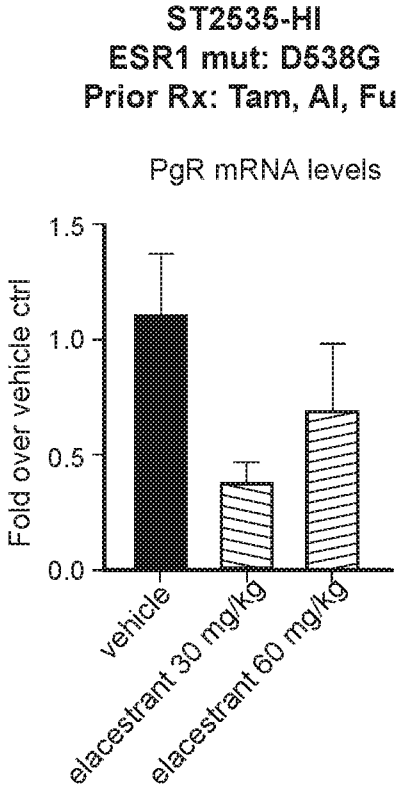

ST2535-HI
ESR1 mut: D538G
Prior Rx: Tam, AI, Ful
PgR mRNA levels
Fold over vehicle ctrl
1.5
1.0
0.5
0.0
vehicle
elacestrant 30 mg/kg
elacestrant 60 mg/kg

FIG. 5A

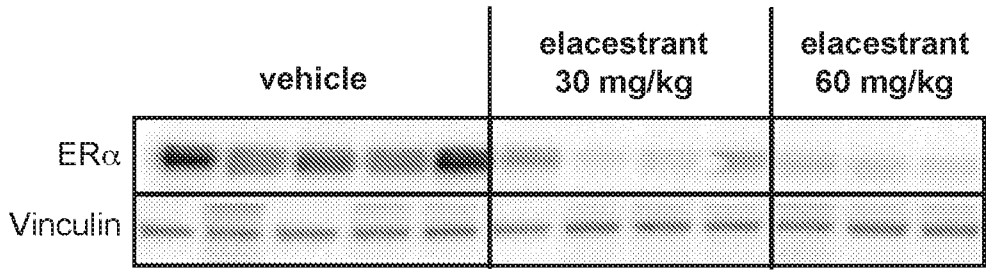

vehicle
elacestrant 30 mg/kg
elacestrant 60 mg/kg
ERα
Vinculin

FIG. 5B

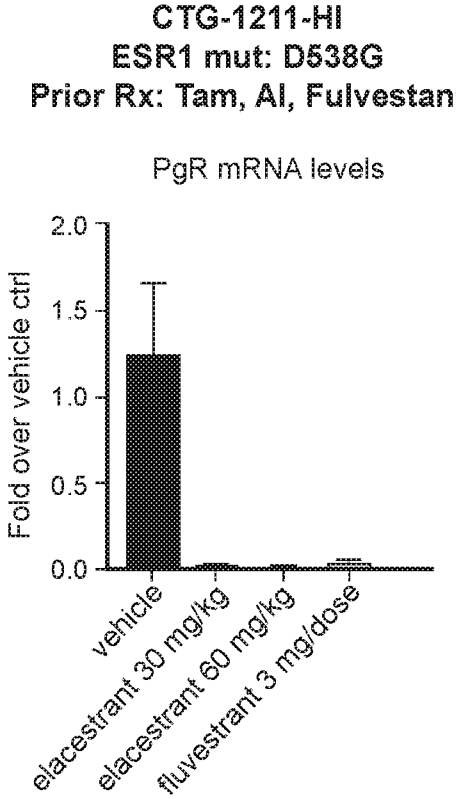

CTG-1211-HI
ESR1 mut: D538G
Prior Rx: Tam, AI, Fulvestant
PgR mRNA levels
Fold over vehicle ctrl
2.0
1.5
1.0
0.5
0.0
vehicle
elacestrant 30 mg/kg
elacestrant 60 mg/kg
fluvestrant 3 mg/dose

FIG. 5C

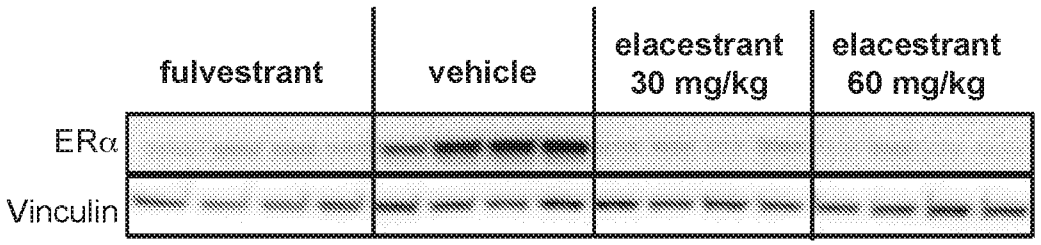

fulvestrant
vehicle
elacestrant 30 mg/kg
elacestrant 60 mg/kg
ERα
Vinculin

FIG. 5D

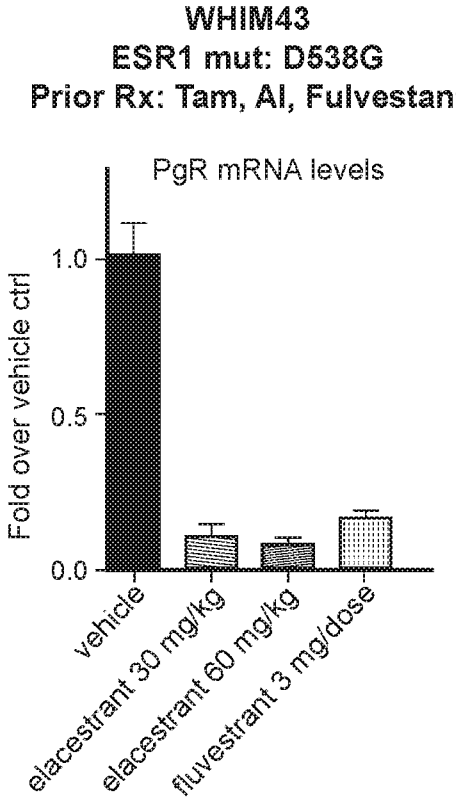

WHIM43
ESR1 mut: D538G
Prior Rx: Tam, AI, Fulvestant
PgR mRNA levels
Fold over vehicle ctrl
1.0
0.5
0.0
vehicle
elacestrant 30 mg/kg
elacestrant 60 mg/kg
fluvestrant 3 mg/dose

FIG. 5E

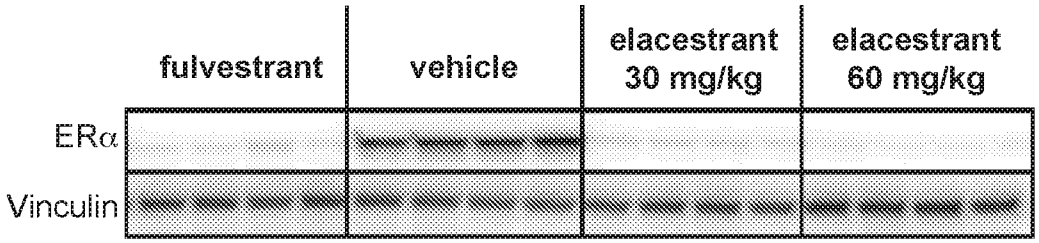

fulvestrant
vehicle
elacestrant 30 mg/kg
elacestrant 60 mg/kg
ERα
Vinculin

FIG. 5F

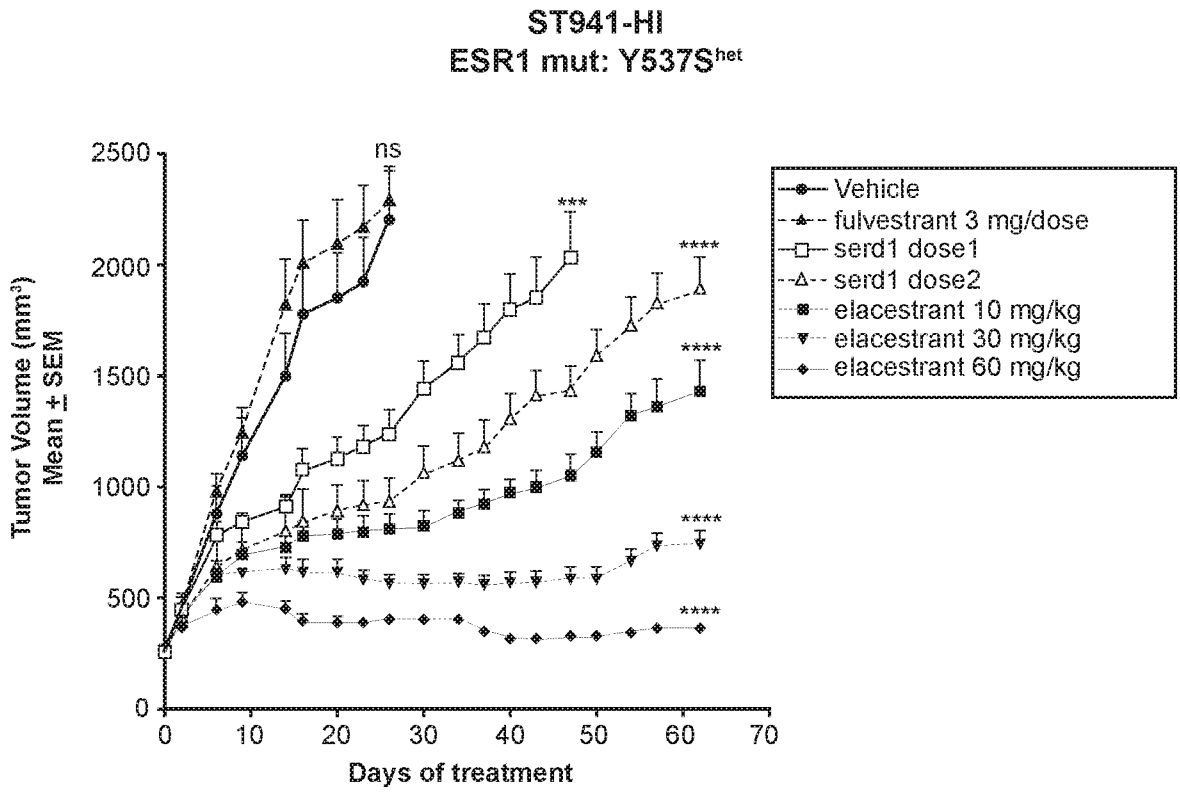

ST941-HI
ESR1 mut: Y537S^het
Tumor Volume (mm³)
Mean ± SEM
Days of treatment
ns
***
****
Vehicle
fulvestrant 3 mg/dose
serd1 dose1
serd1 dose2
elacestrant 10 mg/kg
elacestrant 30 mg/kg
elacestrant 60 mg/kg

FIG. 6A

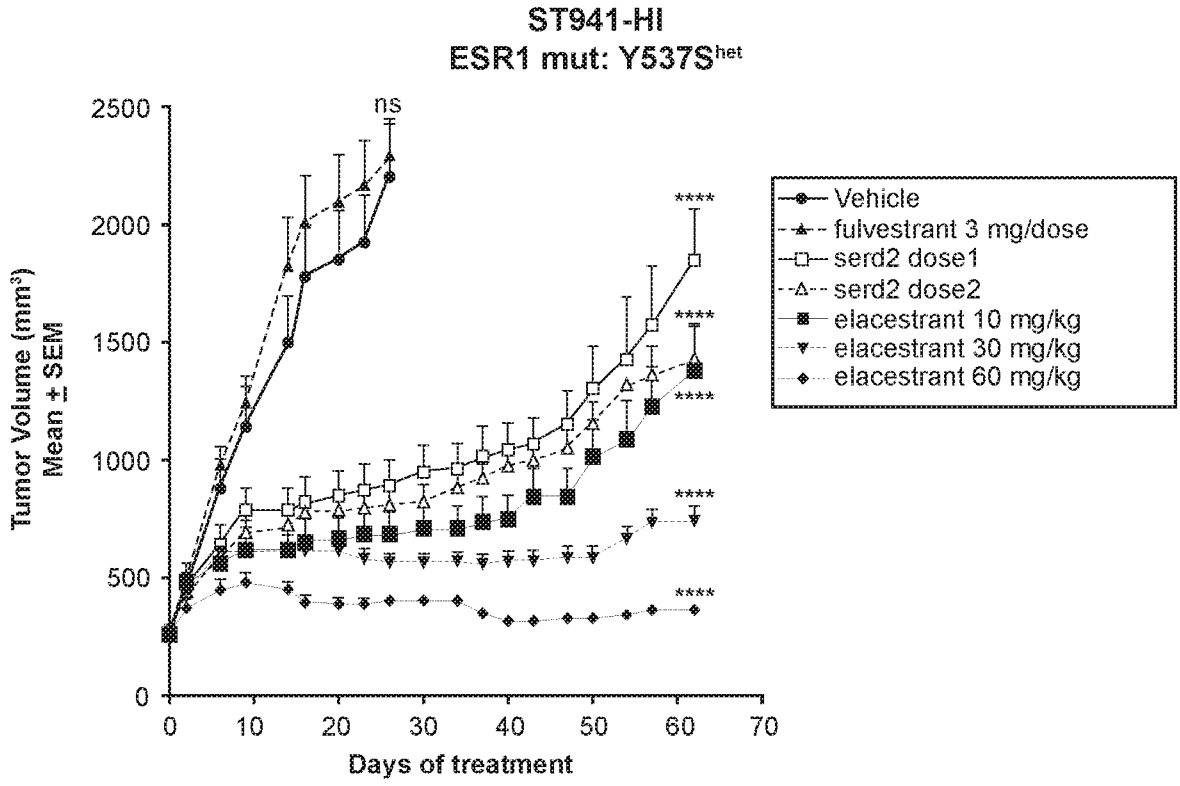

ST941-HI
ESR1 mut: Y537S^het
Tumor Volume (mm³)
Mean ± SEM
Days of treatment
ns
****
Vehicle
fulvestrant 3 mg/dose
serd2 dose1
serd2 dose2
elacestrant 10 mg/kg
elacestrant 30 mg/kg
elacestrant 60 mg/kg

FIG. 6B

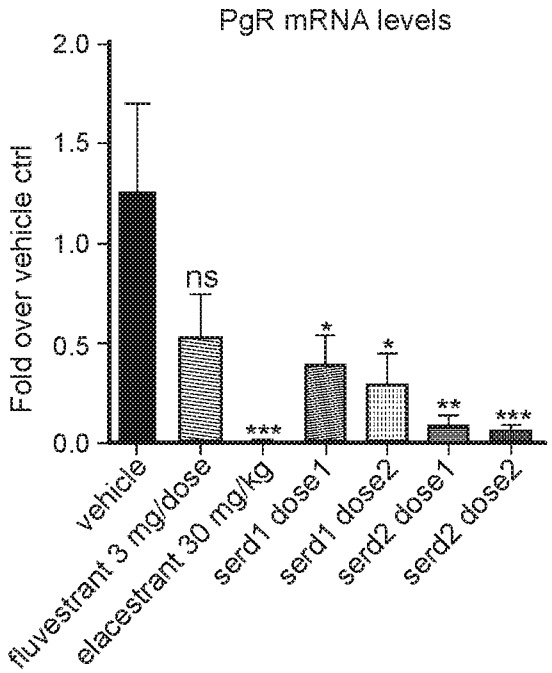


FIG. 7A

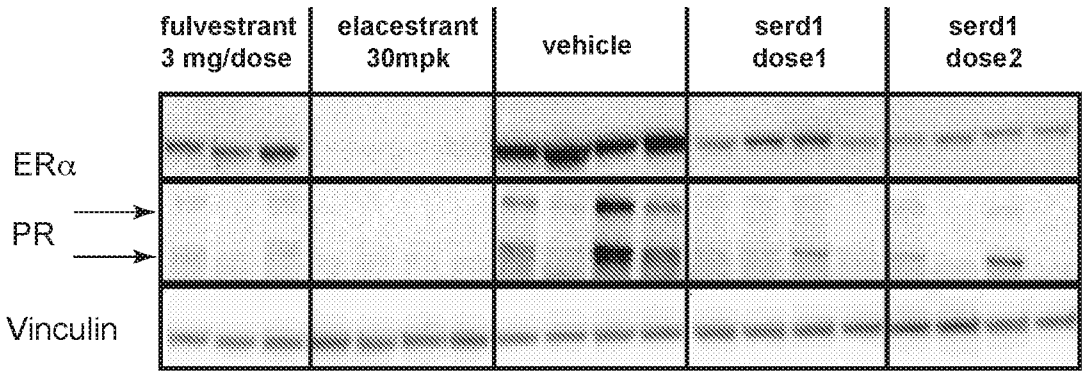


FIG. 7B

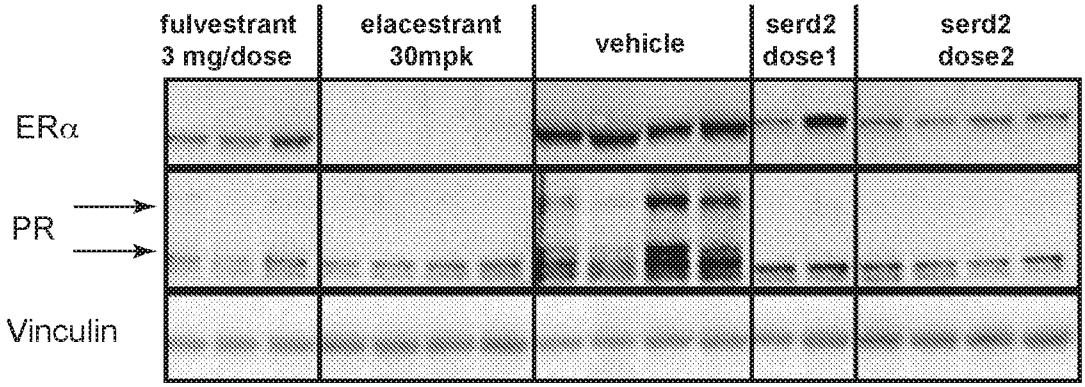


FIG. 7C

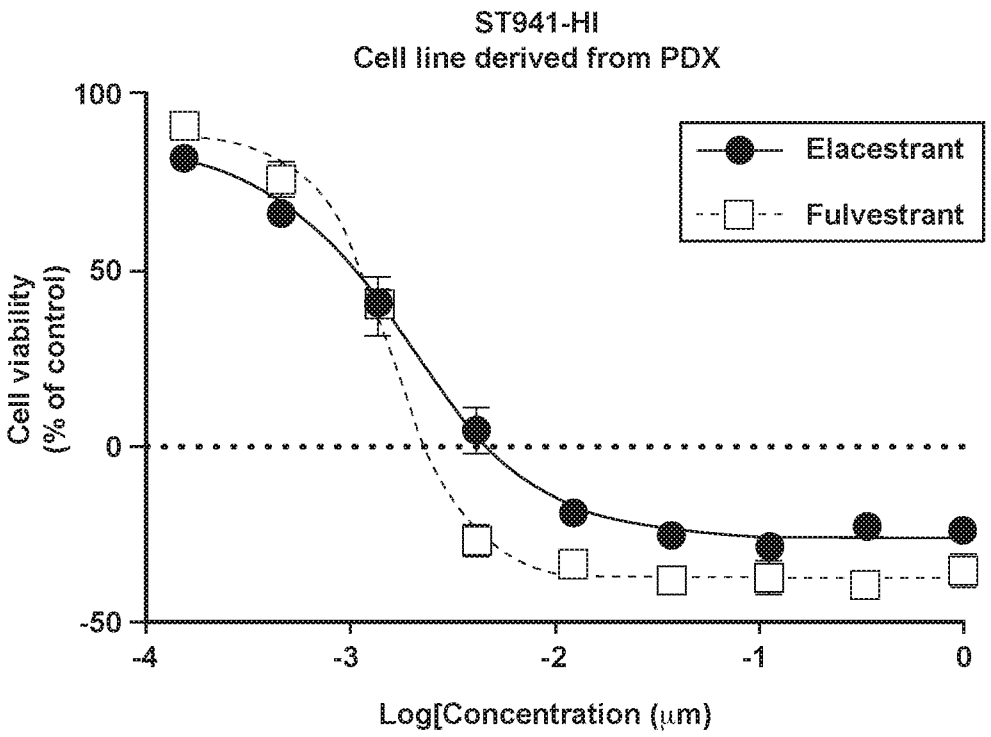

ST941-HI
Cell line derived from PDX
Elacestrant
Fulvestrant
Cell viability
(% of control)
100
50
0
-50
-4
-3
-2
-1
0
Log[Concentration (μm)

FIG. 8A

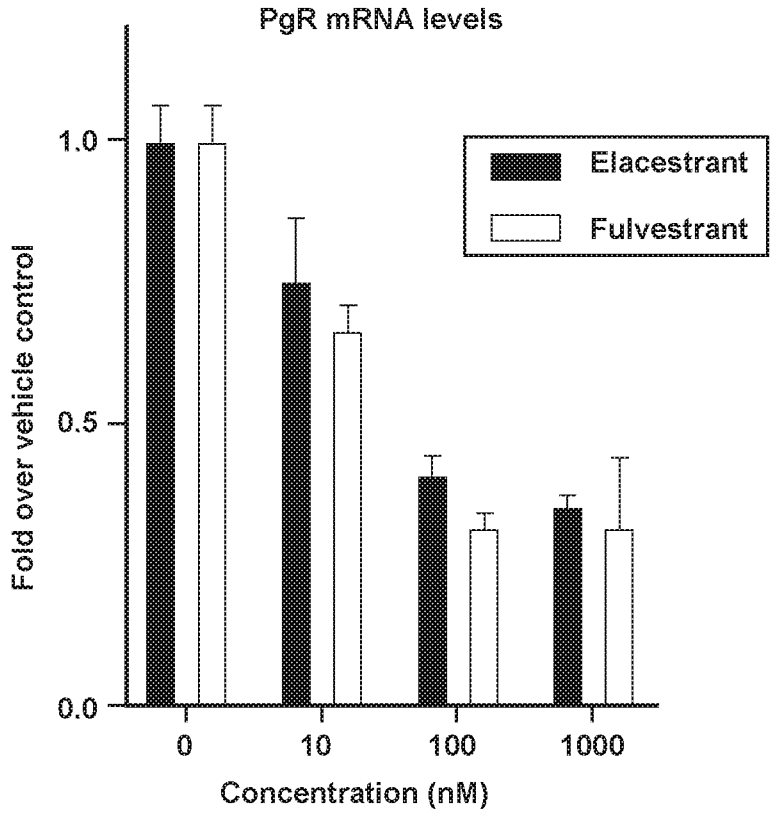

PgR mRNA levels
Elacestrant
Fulvestrant
Fold over vehicle control
1.0
0.5
0.0
0
10
100
1000
Concentration (nM)

FIG. 8B

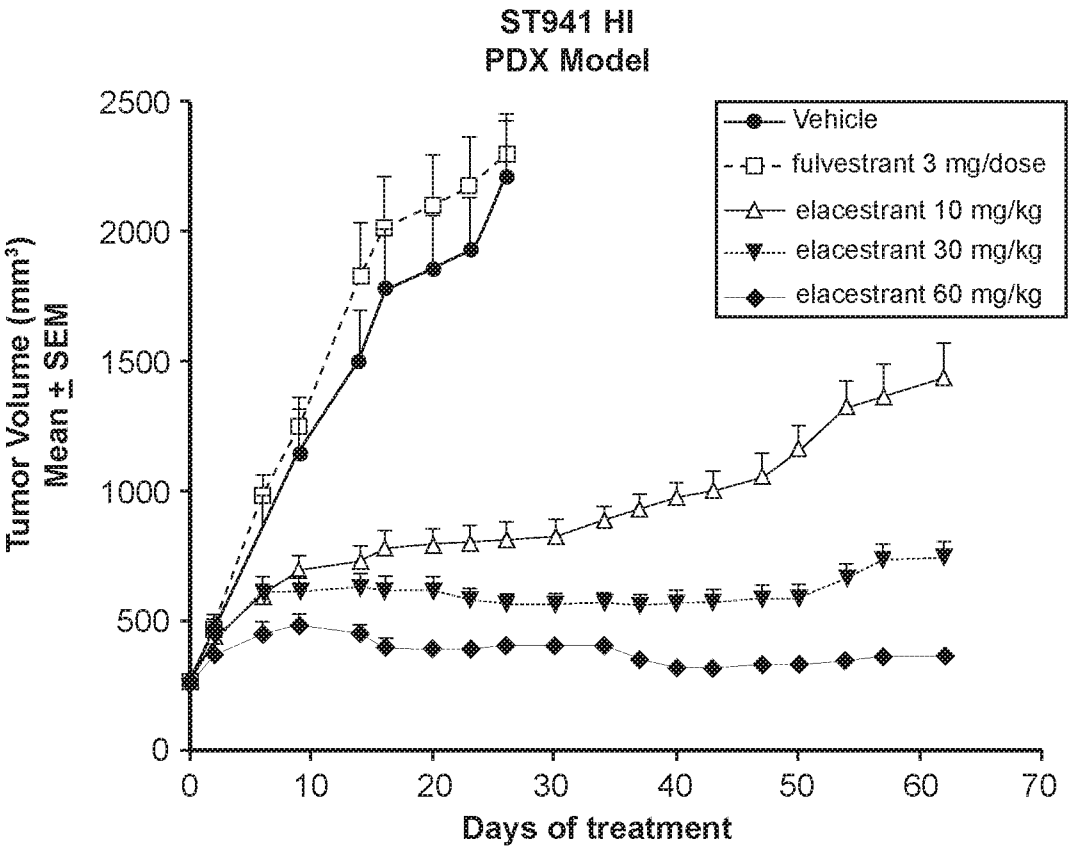

ST941 HI
PDX Model
Tumor Volume (mm³)
Mean ± SEM
2500
2000
1500
1000
500
0
0
10
20
30
40
50
60
70
Days of treatment
Vehicle
fulvestrant 3 mg/dose
elacestrant 10 mg/kg
elacestrant 30 mg/kg
elacestrant 60 mg/kg

FIG. 9A

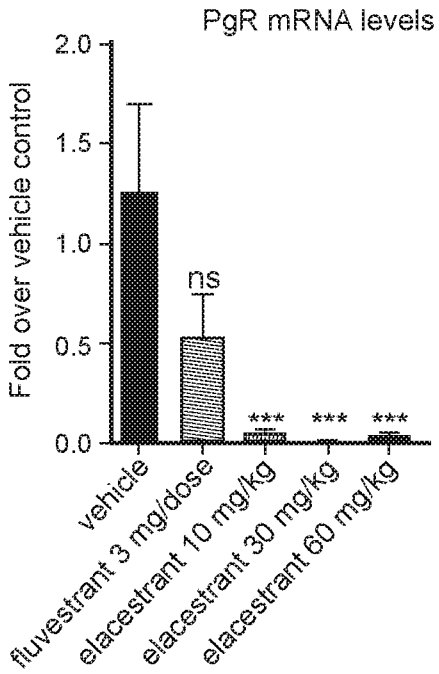
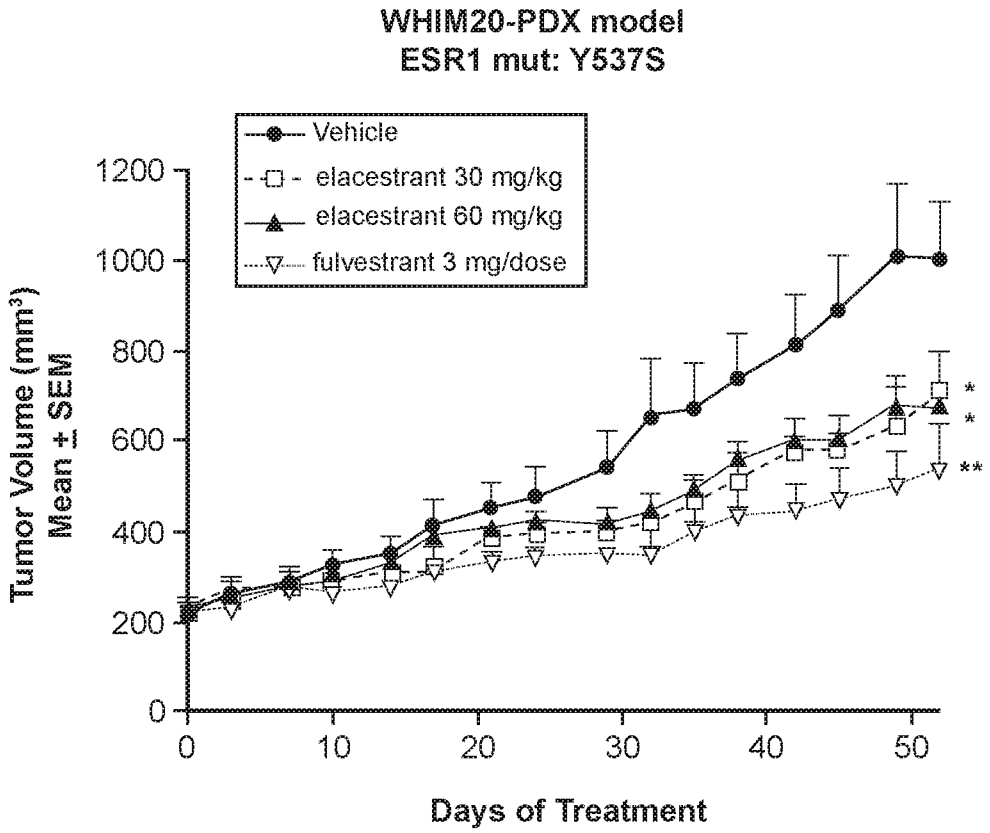

PgR mRNA levels
Fold over vehicle control
2.0
1.5
1.0
0.5
0.0
ns
***
***
***
vehicle
fulvestrant 3 mg/dose
elacestrant 10 mg/kg
elacestrant 30 mg/kg
elacestrant 60 mg/kg

FIG. 9B



ns= not significant, *p <0.05, **p<0.01

FIG. 10A

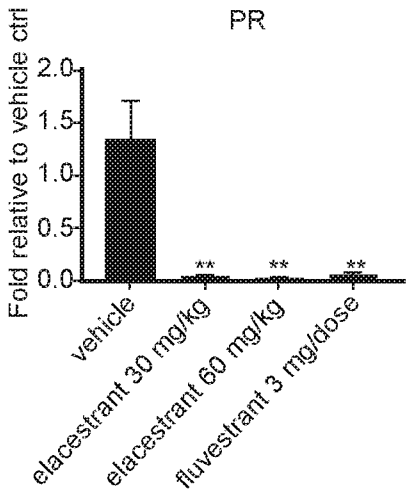


FIG. 10B

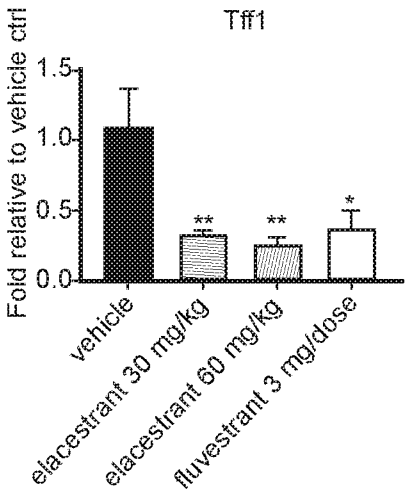


FIG. 10C

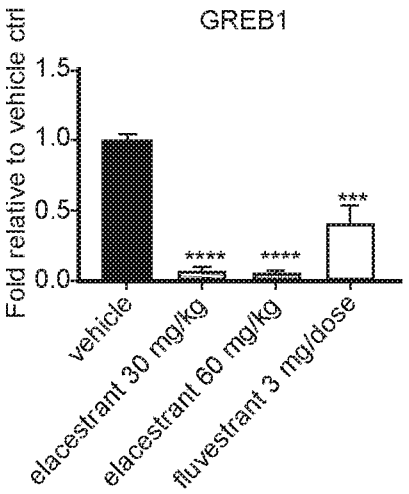


FIG. 10D

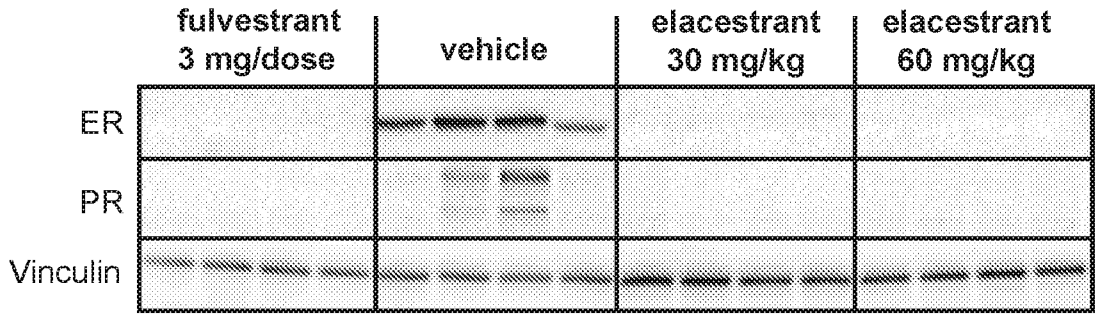

fulvestrant
3 mg/dose
vehicle
elacestrant
30 mg/kg
elacestrant
60 mg/kg
ER
PR
Vinculin

FIG. 10E