Supplementary Information

Supplementary Note

The study protocol is available with the previously published primary analysis (DOI: 10.1001/jamaoncol.2019.3692).

Supplementary Methods

List of additional antibodies used in this study

Antibodies used in assays included pertuzumab rhuMAb 2C4 (supplied by Genentech Inc., lot no. 67966-40/anti2C4907-2, diluted to 100 µg/mL stock solution), biotin-rhuMAb 2C4 (supplied by Genentech Inc., lot no. mehrabak-16Jun17-29, diluted to 600 µg/mL stock solution); DIG-rhuMAb 2C4 (supplied by Genentech Inc., lot no. mehrabak-16Jun17-95, diluted to 600 µg/mL stock solution); peroxidase-conjugated IgG fraction monoclonal mouse anti-digoxin (supplied by Jackson Immuno Research, lot no. 126520, 133869, 140839, diluted to 800 µg/mL stock solution); anti-hu2C4 mAb 4290, Clone 19A2.7 (supplied by Genentech Inc., lot no. ikim-14Aug13-48 [PUR47100], dilutions were variable depending on the assays used); PTEN (138G6) rabbit mAb (supplied by Cell Signaling Technology; catalog no. #9559; dilutions were variable depending on the assays used).

Supplementary Table 1 | Baseline demographics and disease characteristics

Demographic/characteristic	Pertuzumab arm (*n* = 219)	Placebo arm (*n* = 110)
Median age, years (range)	49 (24–72)	49 (27–70)
Age, *n* (%)		
<40 years	40 (18.3)	18 (16.4)
40–49 years	75 (34.2)	40 (36.4)
50–64 years	96 (43.8)	44 (40.0)
≥65 years	8 (3.7)	8 (7.3)
Region, *n* (%)		
Mainland China	175 (79.9)	86 (78.2)
Taiwan	18 (8.2)	13 (11.8)
Other	26 (11.9)	11 (10.0)
ECOG Performance Status, *n* (%)		
0	198 (90.4)	97 (88.2)
1	21 (9.6)	13 (11.8)
Hormone receptor status (IxRS), *n* (%)		
ER- and PgR-negative	105 (47.9)	54 (49.1)
ER- and/or PgR-positive	114 (52.1)	56 (50.9)
Menopausal status, *n* (%)		
Premenopausal	132 (60.3)	65 (59.1)
Postmenopausal	87 (39.7)	45 (40.9)
Disease status (IxRS), *n* (%)		
Early	152 (69.4)	77 (70.0)
Locally advanced	67 (30.6)	33 (30.0)
Primary tumor stage, *n* (%)		
T2	155 (70.8)	71 (64.5)
T3	45 (20.5)	29 (26.4)
T4	19 (8.7)	10 (9.1)
Lymph node status, *n* (%)		
Positive	160 (73.1)	89 (80.9)
Negative	59 (26.9)	21 (19.1)
Histologic subtype,[a] *n* (%)		
Ductal	203 (92.7)	103 (93.6)
Lobular	4 (1.8)	1 (0.9)
Comedo	0	1 (0.9)
Other/comedo	15 (6.8)	8 (7.3)
HER2 IHC score, *n* (%)		
1+	2 (0.9)	0
2+	65 (29.7)	22 (20.0)
3+	152 (69.4)	88 (80.0)
Breast surgery, *n* (%)		
Breast-conserving surgery	57 (26.0)	35 (31.8)
Mastectomy	148 (67.6)	70 (63.6)
Other	5 (2.3)	0
Axillary lymph node surgery, *n* (%)		
Sentinel lymph node biopsy	34 (15.5)	17 (15.5)
Axillary lymph node dissection	171 (78.1)	88 (80.0)
Other	5 (2.3)	0
Adjuvant FEC treatment, *n* (%)	208 (95.0)	103 (93.6)
Adjuvant anti-HER2 treatment, *n* (%)	204 (93.2)	99 (90.0)
Radiotherapy, *n* (%)	154 (70.3)	79 (71.8)

[a]More than one histologic subtype could be selected.

ECOG, Eastern Cooperative Oncology Group; ER, estrogen receptor; FEC, 5-fluorouracil, epirubicin, and cyclophosphamide; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; IxRS, interactive voice/web response system; PgR, progesterone receptor.

Supplementary Table 2 | Exposure to pertuzumab or placebo in the adjuvant treatment period (safety-evaluable population)

	Pertuzumab group (*n* = 208)	Placebo group (*n* = 103)
Treatment duration (weeks)		
n	204	99
Mean (SD)	38.9 (2.4)	37.8 (6.2)
Median	39.0	39.0
Min.–max.	15–43	3–42
Number of cycles		
n	204	99
Mean (SD)	12.9 (0.8)	12.6 (2.0)
Median	13.0	13.0
Min.–max.	5–13	1–13
Number of cycles (group)		
n	204	99
1–12	8 (3.8)	5 (4.9)
13	196 (94.2)	94 (91.3)
>13	0	0
Cumulative dose (mg)		
n	204	99
Mean (SD)	5,828.5 (320.1)	5,693.3 (857.4)
Median	5,880.0	5,880.0
Min.–max.	2,520–6,300	840–6,300
Number of infusion modifications, *n* (%)		
n	204	99
0	202 (97.1)	99 (96.1)
1	2 (1.0)	0
2	0	0
>2	0	0
Number of infusion modifications due to adverse event, *n* (%)		
n	204	99
0	202 (97.1)	99 (96.1)
1	2 (1.0)	0
2	0	0
>2	0	0

SD, standard deviation.

Supplementary Table 3 | Exposure to trastuzumab in the adjuvant treatment period (safety-evaluable population)

	Pertuzumab group (*n* = 208)	Placebo group (*n* = 103)
Treatment duration (weeks)		
n	204	99
Mean (SD)	38.9 (2.4)	37.8 (6.2)
Median	39.0	39.0
Min.–max.	15–43	3–42
Number of cycles		
n	204	99
Mean (SD)	12.9 (0.8)	12.6 (2.0)
Median	13.0	13.0
Min.–max.	5–13	1–13
Number of cycles (group)		
n	204	99
1–12	6 (2.9)	5 (4.9)
13	198 (95.2)	94 (91.3)
>13	0	0
Cumulative dose (mg)		
n	204	99
Mean (SD)	4,847.8 (929.2)	4,601.8 (1049.4)
Median	4720.0	4,720.0
Min.–max.	2,016–7,920	382–6,720
Number of infusion modifications, *n* (%)		
n	204	99
0	196 (94.2)	98 (95.1)
1	8 (3.8)	1 (1.0)
2	0	0
>2	0	0
Number of infusion modifications due to adverse event, *n* (%)		
n	204	99
0	199 (95.7)	98 (95.1)
1	5 (2.4)	1 (1.0)
2	0	0
>2	0	0

SD, standard deviation.

Supplementary Table 4 | Exposure to FEC in the adjuvant treatment period (safety-evaluable population)

	5-fluorouracil		Epirubicin		Cyclophosphamide	
	Pertuzumab group (*n* = 208)	Placebo group (*n* = 103)	Pertuzumab group (*n* = 208)	Placebo group (*n* = 103)	Pertuzumab group (*n* = 208)	Placebo group (*n* = 103)
Treatment duration (weeks)						
n	198	101	208	103	208	103
Mean (SD)	8.8 (1.4)	8.9 (1.3)	9.1 (0.8)	9.1 (0.8)	9.1 (0.8)	9.1 (0.8)
Median	9.0	9.0	9.0	9.0	9.0	9.0
Min.–max.	3–13	3–11	3–13	3–11	3–13	3–11
Number of cycles						
n	198	101	208	103	208	103
Mean (SD)	2.9 (0.4)	2.9 (0.4)	3.0 (0.2)	3.0 (0.2)	3.0 (0.2)	3.0 (0.2)
Median	3.0	3.0	3.0	3.0	3.0	3.0
Min.–max.	1–3	1–3	1–3	1–3	1–3	1–3
Number of cycles (group)						
n	198	101	208	103	208	103
1	7 (3.4)	4 (3.9)	1 (0.5)	1 (1.0)	1 (0.5)	1 (1.0)
2	9 (4.3)	3 (2.9)	2 (1.0)	1 (1.0)	2 (1.0)	1 (1.0)
3	182 (87.5)	94 (91.3)	205 (98.6)	101 (98.1)	205 (98.6)	101 (98.1)
>3	0	0	0	0	0	0
Cumulative dose (mg)						
n	198	101	208	103	208	103
Mean (SD)	2,388.6 (454.9)	2,384.7 (433.2)	447.5 (53.5)	438.8 (52.2)	2,522.1 (343.6)	2,482.9 (328.1)
Median	2,400.0	2,400.0	450.0	442.0	2,475.0	2,475.0
Min.–max.	612–3,564	850–3,150	110–603	170–564	612–3,600	850–3,150
Number of dose modifications, *n* (%)						
n	198	101	208	103	208	103
0	176 (84.6)	96 (93.2)	182 (87.5)	92 (89.3)	171 (82.2)	92 (89.3)
1	21 (10.1)	2 (1.9)	23 (11.1)	8 (7.8)	35 (16.8)	5 (4.9)
2	1 (0.5)	2 (1.9)	3 (1.4)	3 (2.9)	2 (1.0)	5 (4.9)
>2	0	1 (1.0)	0	0	0	1 (1.0)
Number of dose modifications due to AE, *n* (%)						
n	198	101	208	103	208	103
0	182 (87.5)	98 (95.1)	189 (90.9)	95 (92.2)	191 (91.8)	98 (95.1)
1	15 (7.2)	3 (2.9)	17 (8.2)	7 (6.8)	16 (7.7)	5 (4.9)
2	1 (0.5)	0	2 (1.0)	1 (1.0)	1 (0.5)	0
>2	0	0	0	0	0	0

AE, adverse event; FEC, 5-fluorouracil, epirubicin, and cyclophosphamide; SD, standard deviation.

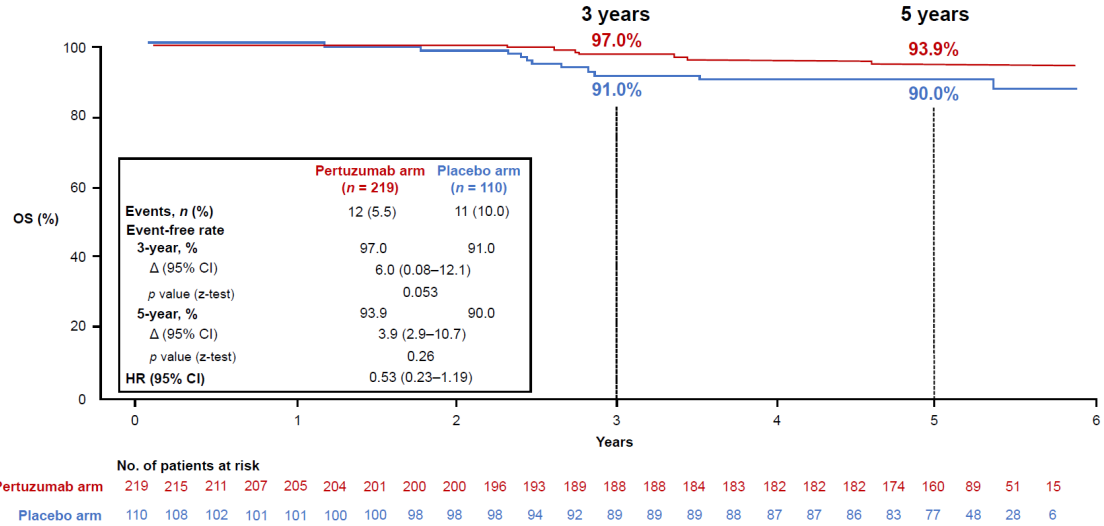
Supplementary Table 5 | Death by cause in the safety-evaluable population

Patients, *n* (%)	**Pertuzumab group (*n* = 218)**	**Placebo group (*n* = 110)**
All deaths	12 (5.5)	11 (10.0)
Other	2 (0.9)	2 (1.8)
Progression of disease	7 (3.2)	5 (4.5)
Recurrence of disease	1 (0.5)	2 (1.8)
Adverse event	2 (0.9)	2 (1.8)
Neoadjuvant study treatment period	1 (0.5)	0
Other	0	0
Progression of disease	0	0
Recurrence of disease	0	0
Adverse event	1 (0.5)	0
Treatment-free follow-up	10 (4.6)	10 (9.1)
Other	2 (0.9)	1 (0.9)
Progression of disease	6 (2.8)	5 (4.5)
Recurrence of disease	1 (0.5)	2 (1.8)
Adverse event	1 (0.5)	2 (1.8)
Death date is missing/wrong	1 (0.5)	1 (0.9)
Other	0	1 (0.9)
Progression of disease	1 (0.5)	0
Recurrence of disease	0	0
Adverse event	0	0

Supplementary Table 6 | List of ethics committees (EC)

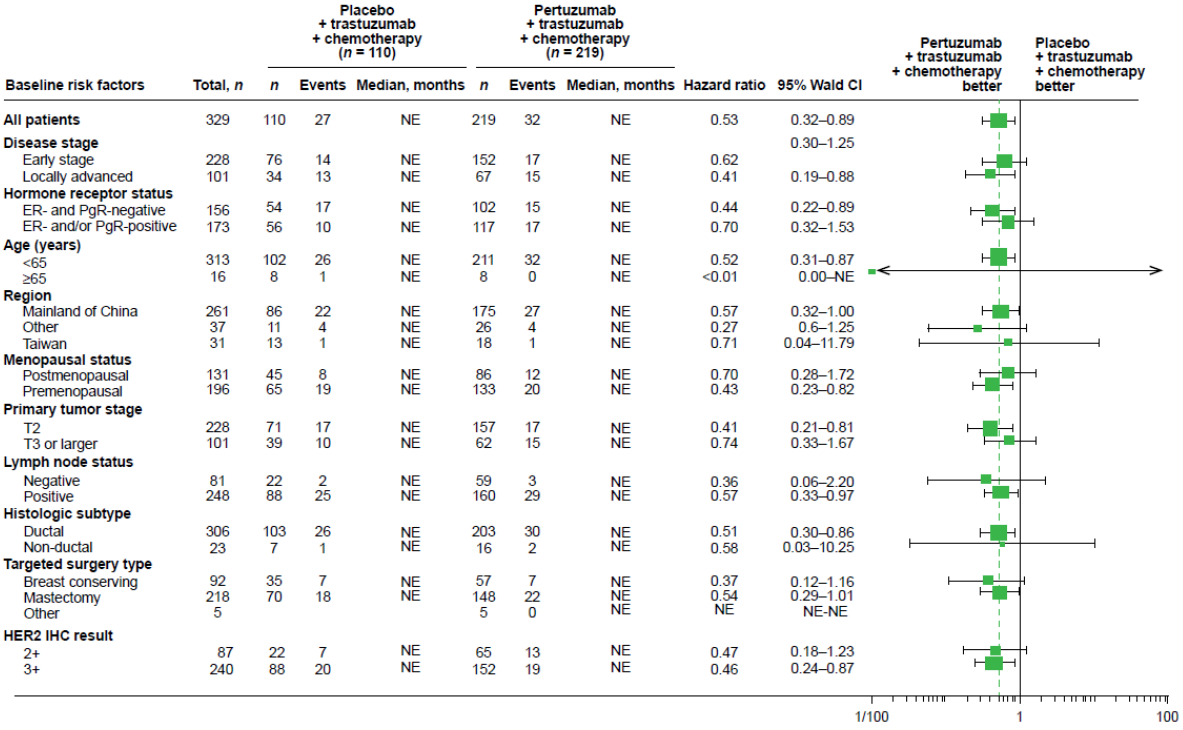
Ethics committees of institutes participating in this study	EC number
EC of Fudan University Shanghai Cancer Center	N/A
EC of The Cancer Hospital of Harbin Medical University	N/A
IEC of Zhejiang Cancer Hospital	N/A
The Ethics Committee of The First Hospital of Jilin University	N/A
EC of Sun Yat-sen University Cancer Center	N/A
Ethics committee of Henan Tumor Hospital	N/A
EC of Guangdong General Hospital	N/A
IEC of Shandong Cancer Hospital	N/A
Fujian Medical University Union Hospital; Ethics Committee	N/A
MacKay Memorial Hospital Institutional Review Board	14CT013b
Research Ethics Committee China Medical University & Hospital	CMUH103-REC2-109
Ajou University Hospital; IRB	N/A
Korea University Guro Hospital; Oncology; IRB	N/A
EC of Ruijin Hospital, Shanghai Jiao Tong University School of Medicine	N/A
Central Research EC of Prince of Songkla University	N/A
307 Hospital Ethics Committee	N/A

Supplementary Fig. 1 | Kaplan–Meier plot of OS in the ITT population.



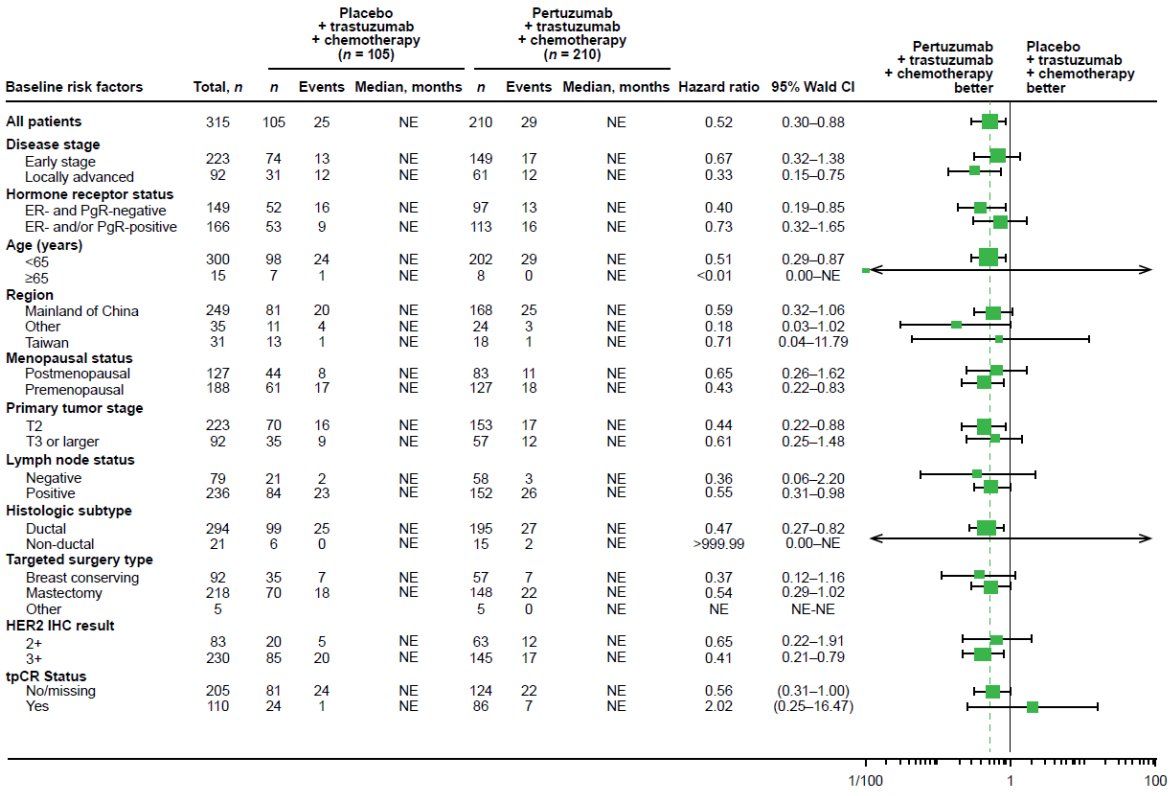
95% CIs and *p* values (two-sided) for differences in event rates were from z-tests using the standard errors for the Kaplan–Meier estimates. CI, confidence interval; HR, hazard ratio; ITT, intention to treat; OS, overall survival.

Supplementary Fig. 2 | Subgroup analysis of EFS according to treatment arm in the ITT population.



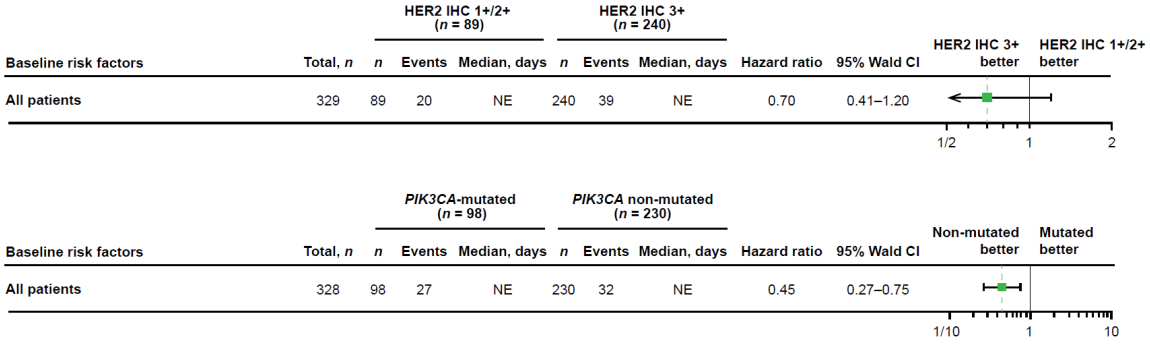
Boxes represent sample sizes and hazard ratios; whiskers represent 95% CI; the vertical dotted line represents the hazard ratio for all patients. CI, confidence interval; EFS, event-free survival; ER, estrogen receptor; FISH, fluorescence *in situ* hybridization; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ITT, intention-to-treat; NE, not evaluable; PgR, progesterone receptor.

Supplementary Fig. 3 | Subgroup analysis of DFS according to treatment arm in the ITT population.



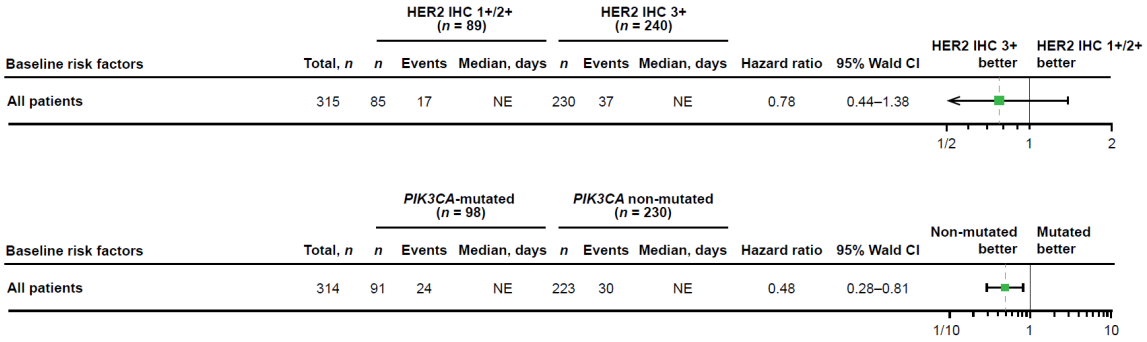
Boxes represent sample sizes and hazard ratios; whiskers represent 95% CI; the vertical dotted line represents the hazard ratio for all patients. CI, confidence interval; DFS, disease-free survival; ER, estrogen receptor; FISH, fluorescence *in situ* hybridization; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ITT, intention-to-treat; NE, not evaluable; PgR, progesterone receptor; tpCR, total pathologic complete response.

Supplementary Fig. 4 | Biomarker subgroup analysis of EFS rates by HER2 IHC and *PIK3CA* baseline biomarker status in the ITT population.



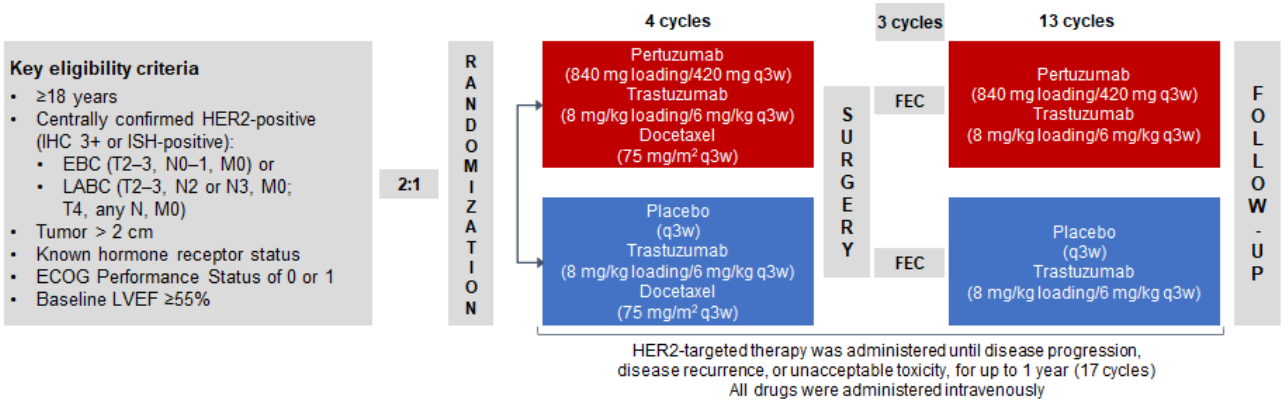
Boxes represent sample sizes and hazard ratios; whiskers represent 95% CI; the vertical dotted line represents the hazard ratio for all patients. CI, confidence interval; EFS, event-free survival; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ITT, intention to treat; NE, not evaluable; *PIK3CA*, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha.

Supplementary Fig. 5 | Biomarker subgroup analysis of DFS rates by HER2 IHC and *PIK3CA* baseline biomarker status in the ITT population.



Boxes represent sample sizes and hazard ratios; whiskers represent 95% CI; the vertical dotted line represents the hazard ratio for all patients. CI, confidence interval; DFS, disease-free survival; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ITT, intention to treat; NE, not evaluable; *PIK3CA*, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha.

Supplementary Fig. 6 | PEONY study design.



EBC, early breast cancer; ECOG, Eastern Cooperative Oncology Group; FEC, 5-fluorouracil, epirubicin, and cyclophosphamide; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridization; LABC, locally advanced breast cancer; LVEF, left ventricular ejection fraction; q3w, every 3 weeks.