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## Obesity increases the incidence of distant metastases in oestrogen receptor-negative human epidermal growth factor receptor 2-positive breast cancer patients

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### KEYWORDS

Body mass index  
Breast neoplasms  
HER2  
Obesity

**Abstract Background:** Obesity is a major negative determinant of breast cancer outcome. However, there are contrasting data on the differential impact of obesity on specific breast cancer subtypes. In particular, very little is known on human epidermal growth factor receptor 2-positive (HER2+) tumours.

**Patients and methods:** We assessed the prognostic role of increased body mass index (BMI) on a consecutive series of non-metastatic HER2+ patients treated at our institution before the introduction of adjuvant Trastuzumab. We separately analysed oestrogen receptor-positive (ER+) and -negative (ER-) HER2+ cases.

**Results:** In ER-/HER2+ tumours we observed a significantly worse overall survival (Hazard ratio (HR) 1.79, *p*-value 0.041) and cumulative incidence of distant metastases (HR 2.03, *p*-value 0.019) in obese (BMI > 30) versus normal/underweight (BMI < 25) patients. Local relapses appeared to be non-significantly reduced in obese patients, masking the overall effect on disease-free survival. Outcome in ER+ tumours, instead, was not significantly different between BMI groups.

**Conclusions:** Obesity significantly correlates with worse overall survival and cumulative incidence of distant metastases in ER-/HER2 positive breast cancer. Differences in the biology

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of breast tumours may determine individual susceptibility to obesity. The biology of the underlying tumour should be taken into account in the design of dietary intervention trials in breast cancer.

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## 1. Introduction

Besides its established role as a risk factor, there is now widespread consensus on the importance of obesity as a negative prognostic factor for breast cancer.<sup>1–3</sup> This recognition has provided a strong rationale for past and on-going studies on the metabolic control of breast cancer patients through dietary or pharmacological interventions.<sup>4–7</sup>

Breast cancer is, however, a biologically heterogeneous disease in which treatment modalities are dictated by molecular features;<sup>8</sup> definitive evidence is still lacking on the impact of obesity on specific breast cancer types. Studies including information on oestrogen receptor (ER)-positive versus -negative tumours were meta-analysed by Niraula et al.<sup>2</sup> The authors concluded that the receptor status did not significantly alter the obesity effect on overall survival (OS) (obese/normal hazard ratio (HR): 1.31 and 1.18 in ER+ and ER–, respectively) or on breast cancer-specific survival (BCSS) (obese/normal HR: 1.36 and 1.46 in ER+ and ER–, respectively). However, there was substantial heterogeneity among the studies included in the analysis, due to differences in body mass index (BMI) categorisation and variability in the definition of the ER– disease, with early studies considering tumours with <10% ER expression as ER negative, contrarily to current guidelines.<sup>8</sup> Individual studies did in fact observe differences between ER+ and ER– groups.<sup>9,10</sup>

If differential effects according to hormone receptor status are still a matter of debate, even less is known on human epidermal growth factor receptor 2-positive (HER2+) tumours. In the present study, we assessed the prognostic role of increased BMI on a consecutive series of non-metastatic HER2+ patients treated at our institution. As the introduction of targeted therapy has radically altered the natural history of this disease, we decided to limit our analysis to patients treated before the introduction of Trastuzumab adjuvant therapy. ER+ and ER– HER2+ tumours were separately analysed, as they constitute clinically and biologically distinct groups.<sup>8,11</sup> Surprisingly, a role for obesity could be observed only in ER– tumours, with a significantly worse OS and an increased risk of distant metastases.

## 2. Patients and methods

We systematically collected information on all consecutive breast cancer patients operated at the European Institute of Oncology in a dedicated data base, thus we

had access to the clinical data of interest (from 1995 to 2005, prior to Trastuzumab adjuvant therapy). We identified 1250 HER2+ early stage breast cancer patients operated during this period for whom data were available regarding weight and height, which we used to calculate the BMI [weight in kilograms/(height in metres)<sup>2</sup>]. Data were also available regarding age, menopausal state, date of surgery, tumour characteristics (histological type, tumour size, nodal involvement, grade, perivascular infiltration, Ki-67 and ER/progesterone receptor (PgR) expression) and treatment modality (type of surgery, adjuvant radiotherapy, endocrine therapy and chemotherapy).

Patients' follow-up included: physical examination every 6 months, annual mammography and breast ultrasound, blood tests every 6–12 months and, in case of symptoms, further evaluation. When possible, the status of those women not attending the scheduled follow-up visits at the institute for more than one year was obtained by telephone contact. Forty patients (3.2%) were lost to follow-up. The median length of follow-up was 8.2 years. The study was approved by the Institutional Review Board.

### 2.1. Statistics

Patients were assigned to BMI groups, according to the standard World Health Organisation (WHO) categorisation,<sup>12</sup> as follows: underweight (BMI < 18.5), normal weight (BMI between 18.5 and 24.99), overweight (BMI between 25 and 29.99) and obese (BMI ≥ 30).

Differences in the distribution of subject characteristics between groups were evaluated by the Chi-square test.

End-points included disease-free survival (DFS), overall survival (OS), cumulative incidence of local or regional recurrence (CI-LR) and distant metastases (CI-DM). DFS was defined as the time from surgery to events such as relapse (including ipsilateral breast recurrence), appearance of a second primary cancer (including contralateral breast cancer), or death, whichever occurred first. OS was defined as the time from surgery until the date of death (from any cause). DFS and OS functions were estimated using the Kaplan–Meier method. The log-rank test was used to assess differences between groups. The CI-LR and CI-DM were defined as the time from surgery to the appearance of a local or regional recurrence and distant metastases, respectively.

CI-LR and CI-DM functions were estimated according to methods described by Kalbfleisch and Prentice,<sup>13</sup>

taking into account the competing causes of recurrence. The Gray's test was used to assess cumulative incidence differences between groups.

The hazard ratios (HRs,) comparing obese patients versus under/normal weight patients and overweight patients versus under/normal weight patients, were estimated using a Cox proportional hazards multivariable model, controlled for age at diagnosis, menopausal status, number of positive lymph nodes, tumour size, grade, percent of oestrogen receptor-positive cells (as a continuous variable), perivascular invasion and type of surgery.

In order to investigate the shape of the relationship between BMI and the hazard of the considered outcomes, multivariable Cox second-order fractional polynomial models were used.<sup>14</sup> Briefly, second-order fractional polynomial models are a family of models generated from power transformations of a continuous exposure variable (in our case BMI), restricted to a predefined set of exponents (in our case the following exponents were tested:  $-2$ ,  $-1$ ,  $-0.5$ ,  $0$ ,  $0.5$ ,  $1$ ,  $2$  and  $3$ ). The best fit among the family of models generated from the combinations of exponents is defined as that with the lowest residual deviance. The family offers considerably flexibility, since a rich set of possible functions, including some 'U-shaped' and 'J-shaped' relations, may be accommodated.

All analyses were carried out with the SAS software (SAS Institute, Cary, NC) and the R software (<http://cran.r-project.org/>) with the *cmprsk* package developed by Gray (<http://biowww.dfci.harvard.edu/~gray/>). All reported *p*-values are two-sided.

### 3. Results

#### 3.1. Study population

Table 1 lists selected demographic, clinical and pathological characteristics and local and systemic treatments of the study population at the time of surgery ( $n = 1250$ ), categorised according to oestrogen receptor (ER) status and WHO BMI classes: normal-underweight (BMI < 25), overweight ( $25 \leq \text{BMI} < 30$ ) and obese (BMI  $\geq 30$ ). Underweight patients were grouped together with normal weight as only 53 patients had a BMI < 18.5.

The prevalence of obese patients in our cohort was 8.1%, in line with the overall prevalence of obesity in the Italian population.<sup>15,16</sup> BMI showed a significantly different distribution ( $p = 0.0032$ ) between the 759 patients with ER positive breast cancer and the 491 patients with ER negative breast cancer (19.5% and 7.8% of overweight and obese patients, respectively, in the ER positive group and 26.7% and 9.6% of overweight and obese patients, respectively, in the ER negative group).

No significant differences were found in either group as regards primary tumour size, lymph node involvement, grade, perivascular invasion or Ki-67 score. Obese and overweight patients were on average significantly older than normal/underweight patients both in the ER positive ( $p < 0.001$ ) and ER negative group ( $p < 0.001$ ). Accordingly, menopausal status was significantly associated with BMI distribution in both groups.

Obese and overweight patients were more likely to have undergone conservative surgery than normal-underweight patients [83.1%, 75.7% and 67%, respectively, in the ER positive group ( $p = 0.009$ ), and 66%, 67.9% and 58.5% in the ER negative group ( $p = 0.142$ )]. No significant differences were noted for adjuvant systemic therapy. However, in the ER positive group, obese patients were more likely to receive anthracycline-containing chemotherapy (22% versus 12% of overweight and normal-underweight patients,  $p = 0.043$ ). None of the patients received Trastuzumab in the adjuvant setting.

#### 3.2. Recurrence in ER+/HER2+ tumours

Overall, 246 first events were observed among the 759 patients with HER2 positive, ER positive breast cancer during the follow-up period (Table 2), with 68 local or regional recurrences, 112 distant metastases and 66 other events (15 contralateral breast cancers, 40 primaries and 11 deaths from causes other than breast cancer). 118 patients died during the follow-up.

No significant differences in OS or DFS could be observed among the three BMI groups. The 5-year OS proportions were 93.4% (95% confidence interval (CI): 91.0–95.2%) in the under/normal weight patients, 94.6% (95% CI: 89.4–97.2%) in the overweight patients and 93.2% (95% CI: 82.9–97.4%) in the obese patients ( $p$ -value 0.849) (Fig. 1A).

The 5-year DFS proportions were 74.8% (95% CI: 70.9–78.2%) in the under/normal weight patients, 76.7% (95% CI: 68.9–82.7%) in the overweight patients and 83.9% (95% CI: 71.2–91.3%) in the obese patients ( $p$ -value 0.554) (Fig. 1B).

We then separated first events into local/regional and distant recurrences and computed the 5-year cumulative incidences. No local recurrence was recorded among obese patients. In contrast, an incidence of 4.1% (95% CI: 1.0–9.9%) and 6.9% (95% CI: 5.0–9.4%) was observed, respectively, in the overweight patients and in the under/normal weight patients; however the differences between the three groups were not statistically significant (Gray test  $p$ -value 0.273) (Fig. 1C). The 5-year CI-DM was comparable in the three groups (Gray test  $p$ -value 0.725) (Fig. 1D).

Table 1  
Baseline characteristics and treatment of patients, by oestrogen receptor status and body mass index (BMI) category.<sup>a</sup>

Variable	ER positive ( $\geq 1\%$ )			<i>p</i> *	ER negative			<i>p</i>
	Under/normal weight <i>N</i> = 552 <i>N</i> (%)	Overweight <i>N</i> = 148 <i>N</i> (%)	Obese <i>N</i> = 59 <i>N</i> (%)		Under/normal weight <i>N</i> = 313 <i>N</i> (%)	Overweight <i>N</i> = 131 <i>N</i> (%)	Obese <i>N</i> = 47 <i>N</i> (%)	
<i>Age at surgery</i>				<0.001				<0.001
<35	49 (8.9)	4 (2.7)	0 (0)		18 (5.8)	2 (1.5)	0 (0)	
35–50	290 (52.5)	40 (27)	19 (32.2)		138 (44.1)	35 (26.7)	11 (23.4)	
51–65	175 (31.7)	84 (56.8)	35 (59.3)		124 (39.6)	73 (55.7)	25 (53.2)	
>65	38 (6.9)	20 (13.5)	5 (8.5)		33 (10.5)	21 (16)	11 (23.4)	
<i>Menopausal status</i>				<0.001				<0.001
Premenopausal	325 (58.9)	48 (32.4)	13 (22)		149 (47.6)	32 (24.4)	10 (21.3)	
Postmenopausal	227 (41.1)	100 (67.6)	46 (78)		164 (52.4)	99 (75.6)	37 (78.7)	
<i>Year of Surgery</i>				0.632				0.890
<1998	4 (0.7)	1 (0.7)	0 (0)		6 (1.9)	2 (1.5)	0 (0)	
1998–2001	206 (37.3)	58 (39.2)	17 (28.8)		120 (38.3)	51 (38.9)	17 (36.2)	
2002–2005	342 (62)	89 (60.1)	42 (71.2)		187 (59.7)	78 (59.5)	30 (63.8)	
<i>Histology</i>				0.829				0.804
Ductal	502 (90.9)	138 (93.2)	52 (88.1)		288 (92)	116 (88.5)	42 (89.4)	
Lobular	19 (3.4)	2 (1.4)	2 (3.4)		4 (1.3)	2 (1.5)	1 (2.1)	
Mixed	13 (2.4)	4 (2.7)	2 (3.4)		0 (0)	0 (0)	0 (0)	
Other	18 (3.3)	4 (2.7)	3 (5.1)		21 (6.7)	13 (9.9)	4 (8.5)	
<i>pT</i>				0.382				0.380
pTX	2 (0.4)	0 (0)	1 (1.7)		4 (1.3)	3 (2.3)	2 (4.3)	
pT1	314 (56.9)	79 (53.4)	26 (44.1)		175 (55.9)	66 (50.4)	20 (42.6)	
pT2	198 (35.9)	60 (40.5)	29 (49.2)		113 (36.1)	47 (35.9)	20 (42.6)	
pT3	32 (5.8)	8 (5.4)	3 (5.1)		17 (5.4)	14 (10.7)	4 (8.5)	
pT4	6 (1.1)	1 (0.7)	0 (0)		4 (1.3)	1 (0.8)	1 (2.1)	
<i>Positive lymph nodes</i>				0.728				0.696
Unknown	11 (2)	4 (2.7)	0 (0)		5 (1.6)	2 (1.5)	0 (0)	
None	283 (51.3)	67 (45.3)	31 (52.5)		159 (50.8)	63 (48.1)	18 (38.3)	
1–3	159 (28.8)	49 (33.1)	19 (32.2)		71 (22.7)	32 (24.4)	13 (27.7)	
4+	99 (17.9)	28 (18.9)	9 (15.3)		78 (24.9)	34 (26)	16 (34)	
<i>Grade</i>				0.311				0.888
Unknown	13 (2.4)	1 (0.7)	3 (5.1)		14 (4.5)	4 (3.1)	1 (2.1)	
1–2	240 (43.5)	65 (43.9)	19 (32.2)		61 (19.5)	26 (19.8)	8 (17)	
3	299 (54.2)	82 (55.4)	37 (62.7)		238 (76)	101 (77.1)	38 (80.9)	
<i>PVI</i>				0.480				0.471
Unknown	1 (0.2)	0 (0)	1 (1.7)		3 (1)	2 (1.5)	2 (4.3)	
Absent	335 (60.7)	94 (63.5)	43 (72.9)		204 (65.2)	81 (61.8)	27 (57.4)	
Present	28 (5.1)	5 (3.4)	2 (3.4)		6 (1.9)	3 (2.3)	1 (2.1)	
Focal	96 (17.4)	27 (18.2)	5 (8.5)		36 (11.5)	24 (18.3)	9 (19.1)	
Diffuse	92 (16.7)	22 (14.9)	8 (13.6)		64 (20.4)	21 (16)	8 (17)	
<i>PgR</i>				0.428				0.621
Unknown	0 (0)	0 (0)	0 (0)		1 (0.3)	0 (0)	0 (0)	
PgR = 0	160 (29)	46 (31.1)	13 (22)		306 (97.8)	129 (98.5)	47 (100)	
PgR > 0	392 (71)	102 (68.9)	46 (78)		6 (1.9)	2 (1.5)	0 (0)	
<i>Ki-67</i>				0.581				0.407
Unknown	2 (0.4)	0 (0)	1 (1.7)		7 (2.2)	4 (3.1)	2 (4.3)	
<14%	48 (8.7)	17 (11.5)	5 (8.5)		11 (3.5)	3 (2.3)	3 (6.4)	
$\geq 14\%$	502 (90.9)	131 (88.5)	53 (89.8)		295 (94.2)	124 (94.7)	42 (89.4)	
<i>Surgery</i>				0.009				0.142
Conservative	370 (67)	112 (75.7)	49 (83.1)		183 (58.5)	89 (67.9)	31 (66)	
Mastectomy	182 (33)	36 (24.3)	10 (16.9)		130 (41.5)	42 (32.1)	16 (34)	
<i>Radiotherapy</i>				0.515				0.542
Unknown	14 (2.5)	7 (4.7)	1 (1.7)		7 (2.2)	7 (5.3)	5 (10.6)	
No	108 (19.6)	28 (18.9)	8 (13.6)		92 (29.4)	31 (23.7)	11 (23.4)	
Yes	430 (77.9)	113 (76.4)	50 (84.7)		214 (68.4)	93 (71)	31 (66)	

(continued on next page)

Table 1 (continued)

Variable	ER positive ( $\geq 1\%$ )			ER negative			<i>p</i>
	Under/normal weight <i>N</i> = 552 <i>N</i> (%)	Overweight <i>N</i> = 148 <i>N</i> (%)	Obese <i>N</i> = 59 <i>N</i> (%)	Under/normal weight <i>N</i> = 313 <i>N</i> (%)	Overweight <i>N</i> = 131 <i>N</i> (%)	Obese <i>N</i> = 47 <i>N</i> (%)	
<i>Adjuvant treatment</i>							0.846
Nil	15 (2.7)	4 (2.7)	1 (1.7)	54 (17.3)	23 (17.6)	6 (12.8)	0.963
Hormonotherapy	157 (28.4)	48 (32.4)	21 (35.6)	1 (0.3)	0 (0)	0 (0)	
Chemotherapy	20 (3.6)	6 (4.1)	1 (1.7)	243 (77.6)	103 (78.6)	39 (83)	
Chemotherapy and hormonotherapy	360 (65.2)	90 (60.8)	36 (61)	15 (4.8)	5 (3.8)	2 (4.3)	
<i>Anthracycline-containing regimen<sup>S</sup></i>							0.043
Yes	305 (82.1)	75 (81.1)	24 (65.0)	190 (75.0)	78 (76.0)	30 (73.0)	0.945
No	67 (18.0)	18 (19.0)	13 (35.0)	65 (25.0)	25 (24.0)	11 (27.0)	

Abbreviations: ER, oestrogen receptors; PgR, progesterone receptors; PVI, peritumoural vascular invasion.

<sup>a</sup> Under/normal weight: BMI < 25; overweight:  $25 \leq \text{BMI} < 30$ ; obese: BMI  $\geq 30$ .

<sup>\*</sup> Chi-square test comparing proportions between BMI categories. *p*-Value were calculated excluding unknown values.

<sup>S</sup> Evaluated only in patients receiving adjuvant chemotherapy.

### 3.3. Recurrence in ER–/HER2+ tumours

Overall, 194 first events were observed among the 491 patients with HER2 positive, ER negative breast cancer during the follow-up period (Table 2), with 52 local or regional recurrences, 104 distant metastases and 38 other events (10 contralateral breast cancers, 14 s primaries and 14 deaths from causes other than breast cancer). 122 patients died during the follow-up.

Within this group of patients, outcome differences between obese patients and the remaining BMI groups were more pronounced. The 5-year OS proportions were 85.9% (95% CI: 81.6–89.3%) in the under/normal weight patients, 82.4% (95% CI: 74.7–87.9%) in the overweight patients, and 73.5% (95% CI: 58.1–84.0%) in the obese patients (*p*-value 0.041) (Fig. 2A).

The 5-year DFS proportions were 69.1% (95% CI: 63.6–73.9%) in the under/normal weight patients, 68.1% (95% CI: 59.3–75.4%) in the overweight patients and 55.2% (95% CI: 39.6–68.4%) in the obese patients (*p*-value 0.170) (Fig. 2B).

Separate analysis of local/regional and distant recurrences revealed a higher 5-year CI-DM in obese patients (33.4%, 95% CI: 22.1–50.5%) than in overweight (17.9%,

95% CI: 12.3–25.9%) and under/normal weight patients (17.5%, 95% CI: 13.8–22.4%). The Gray test *p*-value was 0.019 (Fig. 2D).

Similar to ER+/HER2+ patients, we observed fewer local recurrences in obese patients, with a 5-year CI-LR of 2.3% (95% CI: 0.3–16.0%) versus 7.8% (95% CI: 4.3–14.1%) in the overweight and 10.4% (95% CI: 7.5–14.5%) in the under/normal weight patients. However, the differences between the three groups were not statistically significant (Gray test *p*-value 0.205) (Fig. 2C).

The multivariable analyses (table 3) confirmed the results observed in the univariate analysis: the adjusted risk of death and distant metastases were significantly higher in the obese patients than in the under/normal weight patients (HR for death: 1.79, 95% CI: 1.03–3.10; HR for distant metastases: 2.03, 95% CI: 1.13–3.63), whereas the risk of local/regional events was lower, albeit not statistically significant (adjusted obese versus under/normal weight HR: 0.37, 95% CI: 0.09–1.55).

### 3.4. BMI as a continuous variable

We sought to better characterise the strong association we observed between obesity and distant metastases

Table 2

Observed events, by oestrogen receptor status and body mass index (BMI) category<sup>a</sup>.

Variable	Oestrogen receptor-positive (ER+) ( $\geq 1\%$ )			ER negative (ER–)		
	Under/normal weight <i>N</i> = 552 <i>N</i> (%)	Overweight <i>N</i> = 148 <i>N</i> (%)	Obese <i>N</i> = 59 <i>N</i> (%)	Under/normal weight <i>N</i> = 313 <i>N</i> (%)	Overweight <i>N</i> = 131 <i>N</i> (%)	Obese <i>N</i> = 47 <i>N</i> (%)
Observed first event	184 (33.3)	47 (31.8)	15 (25.4)	121 (38.7)	49 (37.4)	24 (51.1)
Local and regional event	54 (9.8)	12 (8.1)	2 (3.4)	38 (12.1)	12 (9.2)	2 (4.3)
Distant metastases	85 (15.4)	20 (13.5)	7 (11.9)	62 (19.8)	25 (19.1)	17 (36.2)
Other events <sup>b</sup>	45 (8.2)	15 (10.1)	6 (10.2)	21 (6.7)	12 (9.2)	5 (10.6)
Total observed deaths	87 (15.8)	21 (14.2)	10 (16.9)	71 (22.7)	33 (25.2)	18 (38.3)

<sup>a</sup> Under/normal weight: BMI < 25; overweight:  $25 \leq \text{BMI} < 30$ ; obese: BMI  $\geq 30$ .

<sup>b</sup> Including death as first event.



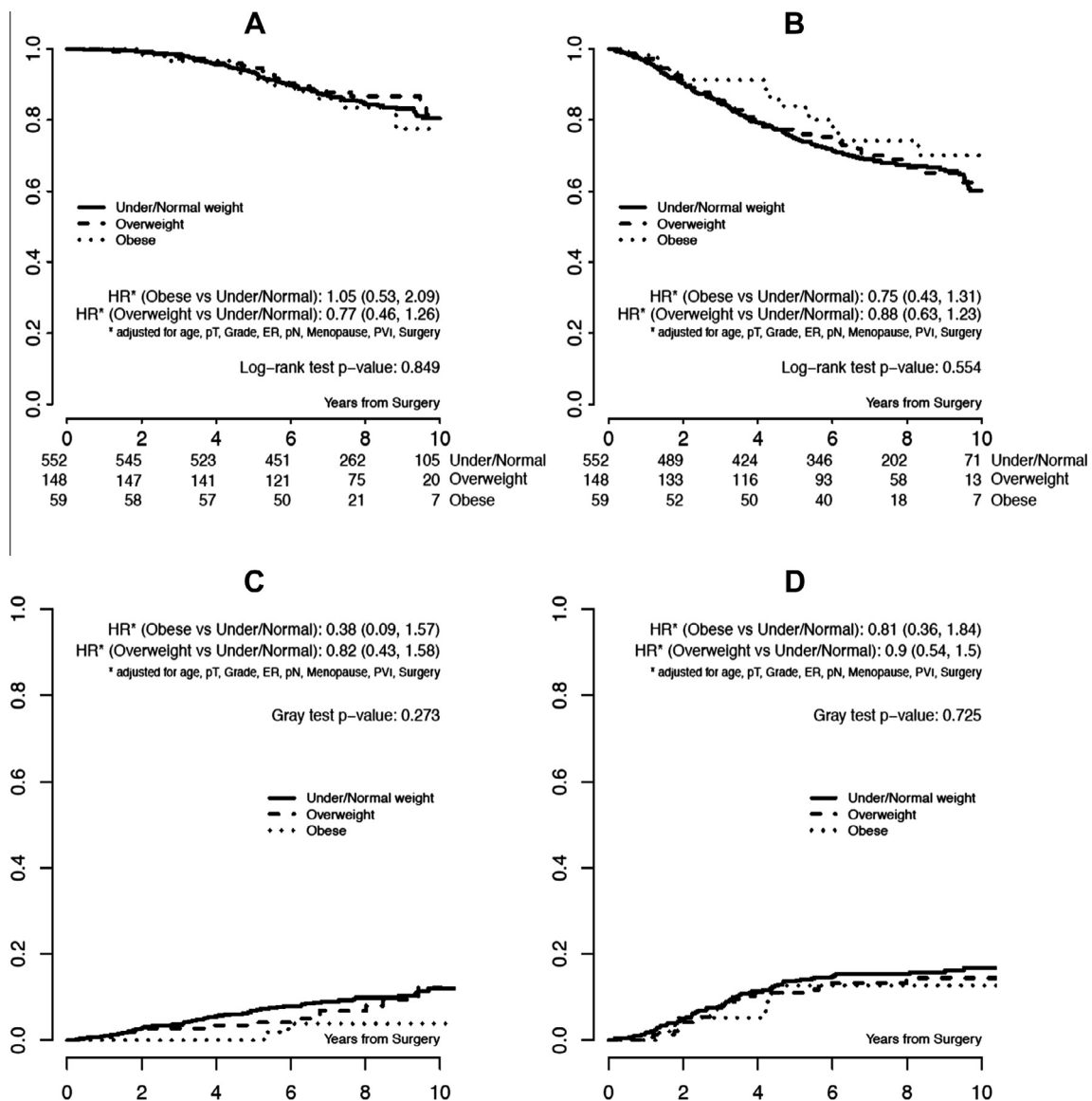


Fig. 1. Overall survival (panel A), disease-free survival (panel B), cumulative incidence of local and regional (LR) recurrence (panel C) and cumulative incidence of distant metastasis (DM) (Panel D), for the 759 patients with oestrogen receptor-positive (ER+)/human epidermal growth factor receptor 2-positive (HER2+) breast cancer, by body mass index category. Univariate log-rank and Gray test  $p$ -values and hazard ratios (HRs) adjusted for age, pT, Grade, ER, pN, Menopause, peritumoural vascular invasion (PVI), Surgery, are reported.

in ER– tumours by treating BMI as a continuous variable, using second-order multivariable fractional polynomial models (Fig. 3).

The relationship between BMI and HR appeared as a U-shaped curve, increasing non-linearly both at high and low BMI values. When we computed the point-wise 95% confidence intervals, the right branch of the curve became significant (lower HR CI bound  $>1.00$ ) at a BMI value of approximately 27.5 (HR: 1.18, 95% CI: 1.01–1.37, when compared with the median BMI value of 24.2), after which the HR continued to grow exponentially with the BMI; on the left side, because of the small number of underweight patients, 95% CIs were much wider and included the 1.00 threshold at any value. The fractional polynomial model fit the data

significantly better than the conventional linear model ( $p = 0.042$ ). Similar analyses with BMI as a continuous variable did not show significant results either in ER+ tumours, or when we studied DFS or the incidence of local events (not shown).

#### 4. Discussion

Little information is available on the impact of BMI on outcome in HER2+ breast cancer.

In this study, we have examined a consecutive series of 1250 patients treated at our institution before 2006, prior to the introduction of adjuvant Trastuzumab. This time frame was chosen in order to provide long term follow-up information and to minimise any potential

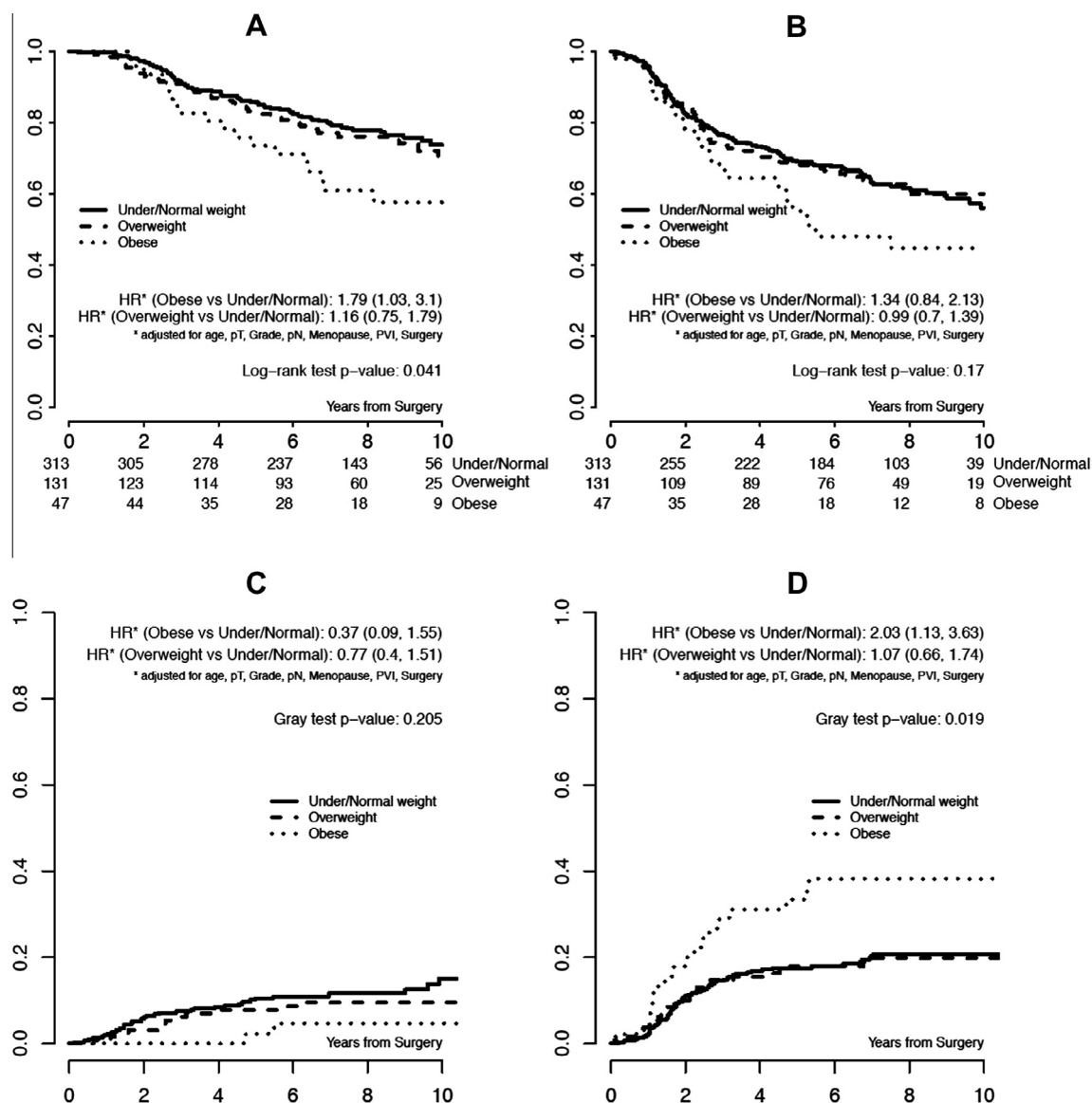


Fig. 2. Overall survival (panel A), disease-free survival (panel B), cumulative incidence of local and regional (LR) recurrence (panel C) and cumulative incidence of distant metastasis (DM) (Panel D), for the 491 patients with oestrogen receptor-negative (ER<sup>-</sup>)/HER2<sup>-</sup> breast cancer, by body mass index category. Univariate log-rank and Gray test *p*-values and hazard ratios (HRs) adjusted for age, pT, Grade, pN, Menopause, peritumoural vascular invasion (PVI), Surgery, are reported.

confounding due to temporal dis-homogeneity in treatment, as the introduction of Trastuzumab radically altered the natural history of this disease. Ongoing retrospective analyses within the Herceptin Adjuvant HERA and the North American Breast Cancer Group trials, for which this study may provide a historical comparison, will address the issue of whether BMI is a predictive factor for patients' response to adjuvant Trastuzumab therapy.

We used the standard WHO BMI categorisation and included our small number of underweight cases into the normal weight group. We stratified patients according to oestrogen receptor status, as this separates two clinically and biologically distinct groups of HER2<sup>+</sup> patients.<sup>8,11</sup>

The main finding of our study is that, within HER2<sup>+</sup> tumours, lack of oestrogen receptor expression separates patients in which obesity correlates with worse outcome from those in which it does not: only ER<sup>-</sup> obese patients had a significantly worse overall survival compared with underweight/normal or overweight patients (HR of 1.79). In the same group the HR for DFS was 1.34 but did not reach statistical significance. However, the number of distant metastases was more than double in obese patients (HR 2.03, *p*-value 0.019), whereas local recurrence showed a slightly lower, albeit not significant, incidence. This pattern is in agreement with previous data on a large unsorted population,<sup>3</sup> and suggests that an apparently modest effect on combined outcome parameters (such as DFS) may be due to the contrasting

Table 3  
Multivariate analysis, by oestrogen receptor status.

		Overall survival (OS)		Disease-free survival (DFS)		Local and regional (LR)		Distant metastasis (DM)	
		Hazard ratio (HR) (95% confidence interval (CI))	<i>p</i>	HR (95% CI)	<i>p</i>	HR (95% CI)	<i>p</i>	HR (95% CI)	<i>p</i>
<i>Oestrogen receptor-positive (ER+) group</i>									
Body mass index (BMI)	Obese versus under/normal weight	1.05 (0.53; 2.09)	0.91	0.75 (0.43; 1.31)	0.31	0.38 (0.09; 1.57)	0.18	0.81 (0.36; 1.84)	0.62
	Overweight versus under/normal weight	0.77 (0.46; 1.26)	0.29	0.88 (0.63; 1.23)	0.45	0.82 (0.43; 1.58)	0.54	0.90 (0.54; 1.50)	0.68
Menopausal status	Pre versus post	1.10 (0.62; 1.95)	0.76	0.99 (0.67; 1.48)	0.97	0.99 (0.45; 2.16)	0.97	0.89 (0.50; 1.60)	0.70
pT	2/3/4 versus 1	1.35 (0.89; 2.06)	0.16	1.31 (0.99; 1.74)	0.06	0.90 (0.52; 1.54)	0.69	1.61 (1.04; 2.48)	0.03
Positive lymph nodes (LN)	1–3 versus none	1.30 (0.79; 2.13)	0.30	0.96 (0.69; 1.34)	0.82	0.80 (0.44; 1.46)	0.47	1.31 (0.78; 2.20)	0.31
	4+ versus none	2.85 (1.74; 4.68)	<0.001	2.10 (1.50; 2.95)	<0.001	1.06 (0.53; 2.13)	0.86	3.37 (2.01; 5.65)	<0.001
Grade	1–2 versus 3	0.58 (0.38; 0.86)	0.01	0.80 (0.61; 1.05)	0.10	0.69 (0.42; 1.15)	0.15	0.69 (0.46; 1.04)	0.08
Peritumoural vascular invasion (PVI)	Present versus absent	1.20 (0.78; 1.83)	0.41	1.28 (0.95; 1.71)	0.10	1.6 (0.94; 2.73)	0.08	1.09 (0.70; 1.69)	0.71
Surgery	Conservative versus mastectomy	0.72 (0.47; 1.09)	0.12	0.84 (0.63; 1.11)	0.22	1.0 (0.57; 1.74)	0.99	0.66 (0.43; 0.99)	0.05
Age	+10 years increase	1.32 (1.03; 1.69)	0.03	1.09 (0.91; 1.29)	0.36	1.0 (0.71; 1.42)	1.00	0.95 (0.74; 1.23)	0.72
<i>ER negative (ER-) group</i>									
BMI	Obese versus under/normal weight	1.79 (1.03; 3.10)	0.04	1.34 (0.84; 2.13)	0.22	0.37 (0.09; 1.55)	0.17	2.03 (1.13; 3.63)	0.02
	Overweight versus under/normal weight	1.16 (0.75; 1.79)	0.50	0.99 (0.70; 1.39)	0.94	0.77 (0.40; 1.51)	0.45	1.07 (0.66; 1.74)	0.78
Menopausal status	Pre versus post	1.13 (0.63; 2.05)	0.68	1.00 (0.63; 1.60)	0.99	2.05 (0.84; 5.02)	0.12	0.74 (0.39; 1.42)	0.37
pT	2/3/4 versus 1	1.57 (1.04; 2.36)	0.03	1.53 (1.11; 2.10)	0.01	1.21 (0.66; 2.23)	0.54	1.62 (1.04; 2.53)	0.03
Positive LN	1–3 versus none	1.73 (1.02; 2.91)	0.04	1.68 (1.14; 2.48)	0.01	1.07 (0.50; 2.32)	0.86	2.62 (1.48; 4.64)	<0.001
	4+ versus none	2.53 (1.52; 4.21)	<0.001	2.75 (1.86; 4.07)	<0.0001	2.26 (1.09; 4.69)	0.03	4.04 (2.27; 7.20)	<0.001
Grade	1–2 versus 3	0.58 (0.32; 1.04)	0.07	0.91 (0.63; 1.32)	0.62	1.11 (0.57; 2.16)	0.77	0.64 (0.36; 1.16)	0.14
PVI	Present versus absent	1.87 (1.21; 2.88)	<0.001	1.22 (0.87; 1.71)	0.24	0.83 (0.41; 1.67)	0.60	1.52 (0.96; 2.39)	0.07
Surgery	Conservative versus mastectomy	1.11 (0.75; 1.64)	0.60	1.45 (1.05; 1.98)	0.02	2.26 (1.15; 4.41)	0.02	1.19 (0.78; 1.82)	0.41
Age	+10 years increase	1.02 (0.78; 1.32)	0.91	0.92 (0.75; 1.14)	0.45	1.28 (0.87; 1.90)	0.21	0.72 (0.53; 0.97)	0.03

association of obesity with local versus distant events. Treating BMI as a continuous variable showed a typical U-shaped correlation with distant metastases, consistent with what observed in previous BMI-cancer mortality studies.<sup>17</sup> Thus, categorising patients according to the standard WHO classification,<sup>12</sup> which is based on cutoff points arbitrarily derived from heterogeneous BMI association studies, may inaccurately reflect the continuous relationship between BMI and cancer outcome variables.

As a whole, outcome was not significantly different between BMI classes in HER2+ tumours, in agreement with the only other study, to date, that separately evaluated HER2+ tumours.<sup>18</sup> No significant differences could be identified within the ER+/HER2+ group. This is consistent with recent data showing that adjuvant endocrine therapy is less effective in obese patients, but only when aromatase inhibitors are used, whereas minimal or no differences are noticed with Tamoxifen.<sup>19,20</sup> Although we do not have detailed information on the



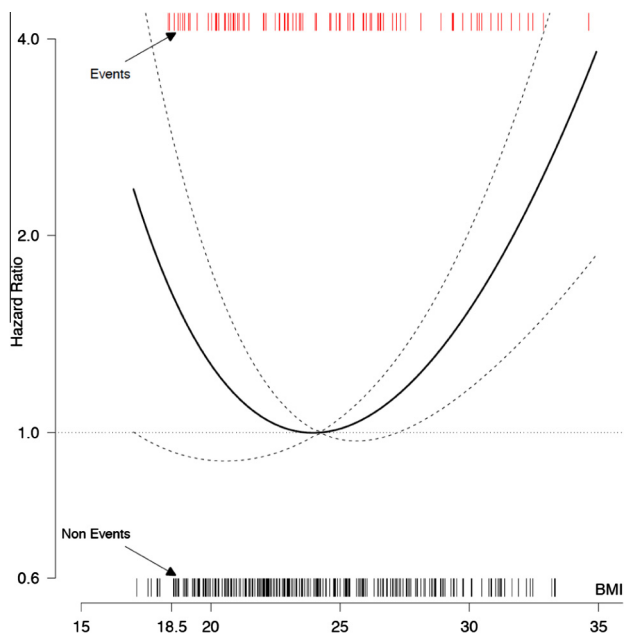


Fig. 3. Relationship between body mass index (BMI), on a continuous scale, and the hazard of distant metastases, in patients with HER2 positive and oestrogen receptor (ER)-negative breast cancer. The relationship is modelled by multivariable fractional polynomials (bold line). Model-based 95% CIs are also reported (dashed lines). The median BMI value (24.2) was considered as the reference value in the estimation of the hazard ratio. Estimates were adjusted for age, pT, Grade, pN, Menopause, peritumoural vascular invasion (PVI), Surgery in a Cox proportional hazard model. The rug plots placed at the top and bottom of the graph show the BMI values in patients that relapsed (top) and in those who did not (bottom).

type of endocrine therapy in our population, given the time window of our analysis it is likely that the vast majority of our patients received Tamoxifen as adjuvant.

Our study has a number of strengths: long median follow-up (8.2 years); detailed treatment history; direct measurement of BMI variables; availability of patient-level data, as opposed to study-level data, which reduces the risk of selection bias and provides results which are closer to everyday clinical practice. Weaknesses are the absence of information on treatment dosage, the retrospective nature of the analysis and the relatively small number of obese patients (8.1%) in the cohort. Such small prevalence of obesity is in line with the general prevalence in Italy at the time,<sup>15</sup> which has only marginally increased in the more recent years and it is significantly smaller than in other Western Countries.<sup>16</sup>

In theory, the poorer outcome of ER–/HER2+ obese patients can be explained by differences in treatment administration and/or biological susceptibility to obesity. Although we do not have information on systemic therapy dosing, treatment variables seem unlikely candidates as: (i) there was no significant difference in the percentage of patients receiving chemotherapy between BMI classes; (ii) the relatively higher frequency

of conservative surgery in obese patients, which is the most evident treatment difference, should more probably affect local recurrence than distant metastases, and this is not the case in our study and (iii) there was no effect in ER+ tumours, which served as a control group in this respect.

We favour a model in which the biology of ER–/HER2+ tumours makes them more prone to metastasisation in response to systemic metabolic alterations. Oestrogen, often invoked as the main mediator of obesity-related progression in breast cancer, does not seem to play a role in this case. Instead, the pattern of relapse described here would suggest an interaction with pathways implicated in metastasis. On a speculative note, one likely candidate in this sense may be the Transforming Growth Factor beta (TGF $\beta$ ) pathway, as suggested by preclinical data showing that (i) increased TGF $\beta$  levels favour metastases in a mouse model of ER–/HER2+ spontaneous tumourigenesis<sup>21</sup> which responds to high fat diet<sup>22</sup> and (ii) TGF $\beta$  plays a crucial role in systemic metabolic control, indeed its circulating levels are dramatically increased in diet-induced or genetic models of obesity.<sup>23</sup> Other mechanisms can also be implicated, such as angiogenesis<sup>21</sup> and inflammatory cytokine imbalance,<sup>24</sup> both of which are perturbed in obese organisms.<sup>25,26</sup>

Our results highlight the importance of the underlying tumour biology in evaluating BMI as a prognostic factor in breast cancer. This may have important consequences for the interpretation and design of dietary intervention trials. For instance, in the Women's Intervention Nutrition Study (WINS), the positive effects of dietary fat reduction on outcome were more evident in ER– tumours,<sup>4</sup> in agreement with our hypothesis that lack of an oestrogen receptor identifies tumours inherently more susceptible to systemic metabolism. However, in the WINS, outcome differences did not reach statistical significance. Stratification for HER2 and ER status could help improve the identification of patients whose cancer prognosis can benefit from weight loss.

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#### Conflict of interest statement

None declared.

#### Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.ejca.2013.07.016>.

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