Circulating HER2/ErbB2 Levels Are Associated With Increased Incidence of Diabetes: A Population-Based Cohort Study

Iram Faqir Muhammad,[1] Yan Borné,[1] Xue Bao,[1,2] Olle Melander,[1] Marju Orho-Melander,[1] Peter M. Nilsson,[1,3] Jan Nilsson,[1] and Gunnar Engström[1]

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OBJECTIVE

HER2/ErbB2 is a member of the epidermal growth factor receptor family. It is widely used as a tumor marker, but it also has recently been associated with insulin resistance. Both ErbB2 and diabetes have been associated with cancer; however, the relationship between ErbB2 and diabetes has not been well explored. The aim of this population-based cohort study was to assess the association between plasma ErbB2 and incidence of diabetes.

RESEARCH DESIGN AND METHODS

The study population included participants from the Malmö Diet and Cancer–Cardiovascular Cohort (age range 46–68 years). After excluding participants with a history of diabetes and those missing data for ErbB2 and other covariates, the final study population consisted of 4,220 individuals. Incidence of diabetes was followed through linkages to local and national registers. Cox proportional hazards regression was used to assess the incidence of diabetes in relation to quartiles of ErbB2, adjusted for potential confounders.

RESULTS

Plasma ErbB2 was significantly and positively associated with glucose, insulin, and HbA_{1c} after being adjusted for potential confounding factors. During a mean ± SD follow-up period of 20.20 ± 5.90 years, 615 participants (14.6%) were diagnosed with new-onset diabetes. Individuals with high levels of ErbB2 had a significantly higher risk of diabetes than those with low levels of ErbB2. The multivariable-adjusted hazard ratio was 1.31 (95% CI 1.03–1.66; $P < 0.05$) for the highest versus the lowest quartile of ErbB2 and was 1.15 (95% CI 1.05–1.25; $P < 0.05$) per 1-SD increase in ErbB2.

CONCLUSIONS

Elevated levels of ErbB2 are associated with increased incidence of diabetes.

The epidermal growth factor receptor (EGFR) proteins are a family of receptor tyrosine kinases that comprises four members: EGFR/ErbB1, ErbB2/HER2/EGFR2/Neu, ErbB3/HER3, and ErbB4/HER4. These kinases are involved in regulating cell growth, survival, and differentiation through multiple signal transduction pathways (1).

The ErbB2/HER2 receptor is encoded by *HER2*, a proto-oncogene located on the long arm of chromosome 17q. No specific ligand for HER2 has been identified, and it relies on heterodimerization with another family member (2), or homodimerization

[1]*Department of Clinical Sciences, Lund University, Malmö, Sweden*
[2]*Nutritional Epidemiology Institute and School of Public Health, Tianjin Medical University, Tianjin, China*
[3]*Department of Internal Medicine, Skåne University Hospital, Malmö, Sweden*

Corresponding author: Iram Faqir Muhammad, iram.faqir_muhammad@med.lu.se

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when expressed at very high levels, to be activated (3). Homo- or heterodimerization leads to autophosphorylation of tyrosine residues within the cytoplasmic domain of the receptors and initiates a variety of downstream signaling pathways. The ErbB2/HER2 oncoprotein consists of three domains: the internal tyrosine portion, which is involved in intracellular signaling; the transmembrane portion; and the external extracellular domain (ECD) (4,5). Proteolytic cleavage results in the release of ECD and the production of an amino terminally truncated cell–associated HER2 fragment with enhanced signaling activity (6). The ECD fragment can be measured in the circulation of women with breast cancer (5). ErbB2 is involved in regulating functions such as differentiation, proliferation, and apoptosis (7), and it plays a vital role in neural and cardiac development (8).

The *HER2* gene is amplified and the protein overexpressed in 15–20% of breast cancers (9,10), and these are associated with a poorer prognosis and a more aggressive phenotype (11,12). HER2 amplification or overexpression has also been observed in a number of other cancers such as ovarian, bladder, salivary gland, endometrial, pancreatic, and non-small-cell lung cancer (13). High circulating levels of ErbB2 have been correlated with amplification and overexpression of the *HER2* gene (14). In short, ErbB2/HER2 in plasma is a specific biomarker for tumors, although slightly raised levels are seen in relation to some nonmalignant conditions such as liver pathologies (15).

Various studies have shown that diabetes is associated with an enhanced risk of cancer (16,17) and all-cause and cancer-related mortality (18,19). Epidemiological evidence links diabetes with an increased risk of certain types of cancer, such as those of the pancreas, liver, bladder, breast, endometrium, colon, and rectum (16). The association between these two conditions may be explained in part by some shared risk factors such as aging, obesity, and diet (16). However, the causes and biological mechanism of this link remain somewhat unclear.

The close relationship observed between diabetes and cancer may support the concept that perturbed metabolism may contribute to the development of oncogenesis. Recent research provides evidence connecting systemic metabolism and cell proliferation, which is one of the key features of cancer (20). Furthermore, in this respect, ErbB2 has shown close associations with molecules governing lipid metabolism (21) and impaired glucose metabolism (22). The notion that glucose metabolism and cancer are associated can also be speculated in relation to the antidiabetes drug metformin, which has reported associations with reduced risk of cancer (23). The anticancer effect of metformin is not clearly understood, but most likely it affects glucose metabolism in the cancer cells. ErbB2 has been widely explored as an oncogenic marker, and it has also been associated with insulin resistance (24). Thus, ErbB2 might have a role beyond oncogenesis. Because of this close association between metabolism and insulin resistance, it might be worthwhile to explore whether a possible link exists between ErbB2 and the risk of developing diabetes, which has not been examined previously.

The aim of this study was to assess the association between circulating levels of ErbB2 in plasma and incidence of diabetes in a population-based cohort.

RESEARCH DESIGN AND METHODS

Study Population

The Malmö Diet and Cancer Study is a prospective study of a population-based cohort from Malmö, a city in the south of Sweden. A random sample of participants (n = 6,103) from this cohort were invited to participate in a substudy, called the Malmö Diet and Cancer–Cardiovascular Cohort, during 1991–1994 (25). A total of 4,721 participants had complete data on ErbB2. Participants with self-reported diabetes or use of antidiabetes medication, or with fasting blood glucose $\geq$6.1 mmol/L (corresponding to a fasting plasma glucose cutoff of 7.0 mmol/L [26]) indicating a diagnosis of diabetes, were excluded. We also excluded participants with missing values for covariates. The final study population consisted of 4,220 participants (mean $\pm$ SD age 57.32 $\pm$ 5.95 years), as depicted in Supplementary Fig. 1. Information regarding HOMA of insulin resistance (HOMA2-IR) and insulin was available for 4,176 participants.

This subcohort was reexamined during 2007–2012, and glucose parameters were measured. Data for fasting glucose were available for 2,817 individuals, and data for glucose 2 h after an oral glucose tolerance test (OGTT) were available for 2,817.

All participants provided written informed consent. The study conformed to the ethical guidelines of the 1975 Declaration of Helsinki and was approved by the Lund University Ethical Committee (LU51/90, LU 2011/537, and LU 2012/762).

Baseline Measurements

The baseline examinations consisted of a self-administered questionnaire, physical examination, and blood sample collection. Information regarding smoking habits and use of antihypertensive medication was obtained from the questionnaire. Smokers were classified into three categories: former smokers, nonsmokers, and current smokers. Blood pressure (millimeters of mercury) was measured once, after 10 min of rest, with a mercury-column sphygmomanometer while the subject was in a supine position. Waist circumference (centimeters) was measured midway between the lowest rib margin and the iliac crest. Standing height was determined by using a fixed stadiometer calibrated in centimeters. Weight (kilograms) was measured by using a calibrated balance-beam scale, with the participants wearing light clothing and no shoes. BMI was calculated as weight (kilograms) divided by the square of height (meters). Blood glucose (millimoles per liter) was determined from blood samples that had been collected while participants were fasting, following standardized procedures at the Department of Clinical Chemistry, Skåne University Hospital. LDL cholesterol was calculated by using Friedewald's formula. Insulin (mIU/L) was determined by using radioimmunoassay. HbA_{1c} (percentage) was determined by using ion exchange chromatography. The HOMA2-IR was calculated with the use of a HOMA2-IR calculator (27). C-reactive protein (CRP; milligrams per liter) was analyzed with a Tina-quant CRP latex assay (Roche Diagnostics, Basel, Switzerland).

The participants in the Malmö Diet and Cancer–Cardiovascular Cohort underwent a reexamination between 2007 and 2012. Fasting plasma glucose was determined by using HemoCue Glucose System (HemoCue AB, Ängelholm, Sweden) (using microcuvette technology) during the reexamination. An OGTT was also

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conducted after an overnight fast, and a repeated plasma glucose measurement was taken 120 min after participants consumed 75 g glucose.

Measurements of ErbB2 and Other Biomarkers

ErbB2 was measured in the plasma from the blood samples acquired during the baseline examinations (1991–1994) and stored at −80°C until the analysis in 2015. The protein was measured with the Olink Proseek Multiplex Oncology I $V2^{96\times96}$ Panel. The values were expressed as normalized protein expression values as arbitrary units on a log2 scale. The lower limit of quantification was 0.95 pg/mL and the upper limit was 15,625 pg/mL. The intra-assay (within a run) coefficient of variation was 5%; the interassay (between runs) value was 21%. The samples were analyzed at the Clinical Biomarkers Facility, Science for Life Laboratory, Uppsala, Sweden. The Olink Proseek Multiplex Oncology I $V2^{96\times96}$ Panel was also used to determine the levels of EGFR, ErbB3/HER3, and ErbB4/HER4 to be used in the sensitivity analysis.

Outcome Ascertainment

All individuals with prevalent diabetes at baseline were excluded from the analyses. The participants were followed from the baseline measurements until the first incidence of diabetes, their emigration from Sweden, their death, or the end of follow-up (31 December 2016), whichever came first. We used both local and national registers to identify incident diabetes cases, which have been explained in detail previously (28). In short, incident diabetes was ascertained from six sources: the Swedish National Diabetes Register, the regional Diabetes 2000 register of the Scania region, the Malmö HbA_{1c} register, the Swedish inpatient register, the Swedish outpatient register, and the nationwide Swedish drug prescription register. In the Swedish National Diabetes Register and the Diabetes 2000 register, new cases of diabetes were diagnosed according to established criteria (fasting plasma glucose concentration ≥7.0 mmol/L resulting from two repeated tests on separate occasions). In the Malmö HbA_{1c} register, subjects were considered to have developed diabetes if they had at least two HbA_{1c} recordings ≥42 mmol/mol (6.0%), based on the Swedish Mono-S-based standardization system (corresponding to 53 mmol/mol [7.0%], according to the U.S. National Glycohemoglobin Standardization Program). Diabetes was diagnosed by a senior physician using the Swedish inpatient and outpatient registers. In the nationwide prescription register, a filled prescription of insulin or antidiabetes medication (Anatomical Therapeutic Chemical Classification System code A10) was required for a diagnosis of diabetes.

Information about the type of diabetes was not available for all 615 incident cases. Of these, 287 (46.7%) were classified as type 2, 15 (2.4%) as type 1, 1 (0.1%) as latent autoimmune diabetes in adults (LADA), and 1 (0.1%) as other type. Information for type of diabetes was lacking for 311 participants (50.6%). In an effort to classify type of diabetes further, we obtained information regarding medication use from the drug prescription register. Those who had been prescribed insulin within 3 years of diagnosis (29) were classified as type 1 (seven participants). This group, together with participants with known type 1 diabetes or LADA, was excluded from a sensitivity analysis.

Statistical Analysis

Because of their skewed distribution, CRP, HOMA2-IR, insulin at baseline, and fasting glucose and 2-h glucose from reexamination were natural log transformed. Participants were categorized into quartiles according to concentration of ErbB2. The characteristics of the study population across the quartiles of ErbB2 were described as mean ± SD (for normal distributions), median (25th–75th percentiles) (for skewed distributions), or percentages. We tested differences across the quartiles using χ^2 for categorical variables and ANOVA for continuous variables. We used multiple linear regression to assess the association between ErbB2 (as the dependent variable) and baseline fasting glucose, HbA_{1c}, HOMA2-IR, and insulin; these were adjusted for potential confounding factors in two separate models. We also assessed the association for fasting glucose and 2-h glucose from the reexamination (2007–2012) in both models.

Analysis was conducted using Cox proportional hazards regression to compare the incidence of diabetes across ErbB2 quartiles. In addition, we calculated the hazard ratio (HR) per 1-SD increment of ErbB2. The SD of ErbB2 was calculated separately for each batch in order to correct for potential batch effects. We calculated HRs with 95% CIs, using the lowest quartile as the reference category. We used time until death, emigration, incident diabetes, or the end of follow-up as the time scale. We constructed four Cox models: Model 1 analyzed the unadjusted association between ErbB2 and incidence of diabetes. Model 2 was adjusted for age, sex, waist circumference, smoking habit, LDL cholesterol, systolic blood pressure, and use of antihypertensive medication. Model 3 was adjusted for model 2 variables and CRP levels. Finally, model 4 was further adjusted for baseline fasting glucose; this model could therefore be regarded as an overadjusted model. As an additional exploration, we used BMI instead of waist circumference in the final model in order to assess the association between ErbB2 levels and incident diabetes. We tested the proportional hazard assumptions by incorporating the time-dependent effects of covariates. Possible interactions between ErbB2 levels and risk factors such as age, sex, glucose, and CRP were investigated by introducing interaction terms in the final multivariate model. We used a Kaplan-Meier curve to illustrate incidence of diabetes in relation to ErbB2 quartiles.

We also assessed correlations between ErbB2 and other members of the EGFR family. In a sensitivity analysis, we further adjusted the association between ErbB2 and incident diabetes for EGFR, ErbB3, and ErbB4 (one at a time) in the final model. Furthermore, we analyzed the association between ErbB2 and incident diabetes after excluding those participants who had been classified as having type 1 diabetes or LADA (n = 24).

A P value <0.05 was regarded as statistically significant. All analyses were performed using SPSS Statistics version 24 (IBM Corp., Armonk, NY).

RESULTS

Baseline Characteristics

The characteristics of the study participants across the quartiles of ErbB2 are presented in Table 1. Participants with elevated levels of ErbB2 generally had higher values of risk factors than those with lower levels. Baseline fasting

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Table 1—Relationships of risk factors across ErbB2 quartiles (n = 4,220)

	ErbB2				
	Quartile 1	Quartile 2	Quartile 3	Quartile 4	*P* value
Participants, *n*	1,055	1,055	1,055	1,055	
ErbB2, AU*	7.33 ± 0.26	7.75 ± 0.07	7.98 ± 0.07	8.33(± 0.18	
CRP, mg/L	1.20 (0.60–2.40)	1.10 (0.60–2.30)	1.30 (0.60–2.60)	1.50 (0.70–3.10)	<0.001
Age, years	56.42 ± 5.94	57.48 ± 5.92	57.60 ± 5.94	57.79 ± 5.94	<0.001
Male sex	265 (25.1)	379 (35.9)	441 (41.8	539 (51.1)	<0.001
Waist circumference, cm	78.60 ± 10.88	81.40 ± 11.94	83.14 ± 12.08	86.44 ± 12.39	<0.001
BMI, kg/m^2	24.70 ± 3.53	25.24 ± 3.68	25.56 ± 3.68	26.23 ± 3.82	<0.001
LDL, mmol/L	3.99 ± 0.96	4.11 ± 0.97	4.20 ± 0.96	4.36 ± 0.99	<0.001
Fasting glucose, mmol/L	4.75 ± 0.42	4.87 ± 0.43	4.89 ± 0.44	5.01 ± 0.44	<0.001
HbA_{1c}, %	4.68 ± 0.40	4.78 ± 0.39	4.81 ± 0.42	4.85 ± 0.44	<0.001
HbA_{1c}, mmol/mol	28	29	29	29	<0.001
HOMA2-IR (*n* = 4,176)	0.70 (0.50–0.90)	0.80 (0.50–1.10)	0.80 (0.50–1.2)	1.0 (0.70–1.50)	<0.001
Insulin, mIU/L (*n* = 4,176)	5.0 (4.0–7.0)	6.0 (4.0–8.0)	6.0 (4.0–9.0)	7.0 (5.0–11.0)	<0.001
Systolic blood pressure, mmHg	135.77 ± 17.78	139.29 ± 18.41	141.07 ± 18.70	144.50 ± 18.54	<0.001
BP-lowering medication	128 (12.1)	150 (14.2)	150 (14.2)	176 (16.7)	0.030
Smoking habit					0.048
Never smoker	465 (44.1)	447 (42.4)	422 (40)	396 (37.5)	
Former smoker	385 (36.5)	376 (35.6)	404 (38.3)	408 (38.7)	
Current smoker	205 (19.4)	232 (22)	229 (21.7)	251 (23.8)	
Reexamination (2007–2012)					
Fasting glucose, mmol/L (*n* = 2,817)	5.7 (5.3–6.1)	5.8 (5.4–6.3)	5.8 (5.4–6.3)	6.0 (5.5–6.6)	<0.001
Glucose 2-h after OGTT, mmol/L (*n* = 2,637)	6.5 (5.4–7.9)	6.6 (5.4–8.0)	6.7 (5.4–8.1)	7.3 (6.0–9.0)	<0.001

Values are the mean ± SD, *n* (%), or median (25th–75th percentiles) unless specified otherwise. AU, arbitrary units; BP, blood pressure. *Normalized protein expression values on a log2 scale.

glucose, HbA_{1c}, HOMA2-IR, and insulin were all positively and significantly correlated with ErbB2 level after adjusting for confounding factors, as shown in Table 2. Similarly, we observed a significantly positive correlation between ErbB2 level and fasting glucose and 2-h glucose at reexamination, which represents a longitudinal association.

Incidence of Diabetes

During a mean ± SD follow-up period of 20.20 ± 5.90 years, 615 participants developed diabetes. The incidence of diabetes was 7.22 per 1,000 person-years in the study population. The diabetes-free survival of the study population in relation to the different quartiles of ErbB2 is presented in Fig. 1.

Analysis using the unadjusted Cox model demonstrated that the participants in the highest quartile had a significantly higher risk of developing diabetes than did those in the lowest quartile (HR 2.41 [95% CI 1.92–3.0]) (Table 3). The risk remained significantly higher when adjusted for covariates in model 2 (HR 1.72 [95% CI 1.36–2.18]). When we adjusted further for CRP, the HR was 1.73 (95% CI 1.37–2.19). The HR was attenuated but was still statistically significant when we then adjusted for baseline fasting glucose (HR 1.31 [95% CI 1.03–1.66]). When we included BMI instead of waist circumference in the final model, the results were similar (HR 1.34 [95% CI 1.06–1.70]). The multivariable-adjusted HR was 1.15 (95% CI 1.05–1.25) for diabetes per 1-SD increment of ErbB2 in the final model. A Kaplan-Meier curve indicated that those with higher levels of ErbB2 had shorter diabetes-free survival, as shown in Fig. 1.

We observed no interaction between ErbB2 and sex, age, baseline fasting glucose, or CRP when we analyzed incident diabetes. The HR per 1-SD increment of ErbB2 was 1.27 (95% CI 1.10–1.46) in men and 1.26 (95% CI 1.11–1.42) in women after adjusting for model 3 variables (*P* for interaction = 0.848). The HR among those aged ≤58 years was 1.34 (95% CI 1.19–1.51) and for those >58 years old was 1.17 (95% CI 1.02–1.35) (*P* for interaction = 0.289).

Table 2—Correlations between ErbB2 and fasting glucose, HbA_{1c}, HOMA2-IR, and insulin at baseline and between fasting glucose and OGTT at reexamination (2007–2012)

	Baseline				Reexamination	
	Fasting glucose (baseline) (*n* = 4,220)	HbA_{1c} (*n* = 4,220)	HOMA2-IR (*n* = 4,176)	Insulin (*n* = 4,176)	Fasting glucose (*n* = 2,817)	OGTT (*n* = 2,637)
Model 1*	0.181***	0.157***	0.231***	0.225***	0.124***	0.109***
Model 2**	0.130***	0.127***	0.202***	0.195***	0.083***	0.078***

Values are standardized β coefficients from multiple linear regressions with ErbB2 as the dependent variable. Natural log-transformed values were used for HOMA2-IR, insulin, CRP, fasting glucose (at reexamination), and OGTT (at reexamination).*Model 1 was adjusted for age and sex. **Model 2 was adjusted for age, sex, waist circumference, smoking habits, systolic blood pressure, LDL cholesterol, use of antihypertensive medication, and CRP. ***$P < 0.001$.

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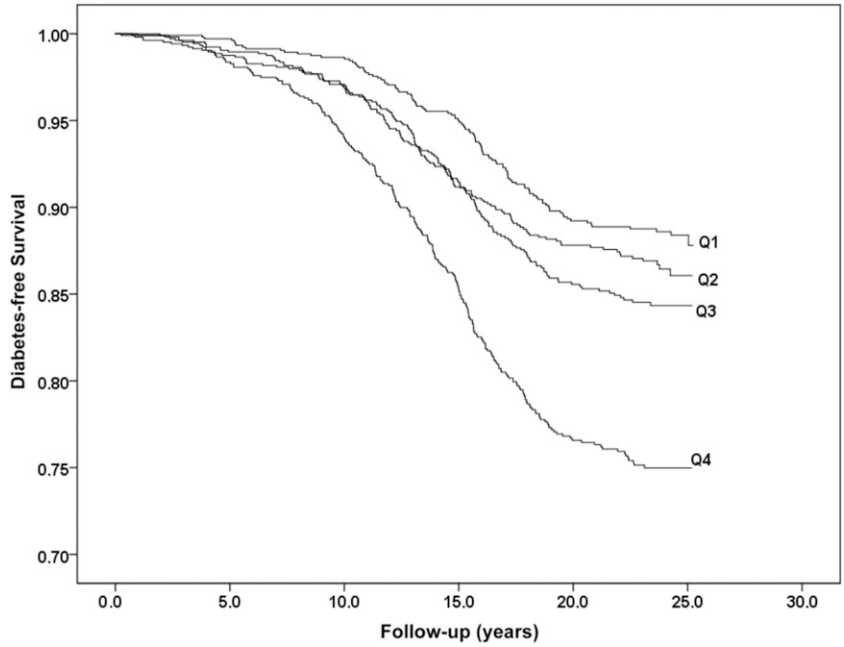

Figure 1—Diabetes-free survival in relation to ErbB2 quartiles (Q).

Sensitivity Analysis

ErbB2 was strongly correlated with other members of the EGFR family (i.e., EGFR, ErbB3, and ErbB4), as shown in Supplementary Table 1. Because ErbB2 forms dimers with other members of the family and they were strongly correlated, it was worth exploring whether adjusting for these proteins changes the association. Therefore, in the sensitivity analysis, we adjusted further by introducing EGFR, ErbB3, and ErbB4 one by one into the final multivariate Cox model. The association between ErbB2 and diabetes remained significant (data not shown).

As part of the sensitivity analysis, we excluded from the study population incident cases of known type 1 diabetes and LADA and participants receiving insulin treatment during the first 3 years after diagnosis. We assessed the association between ErbB2 and type 2 diabetes (confirmed or probable; 591 cases of incident diabetes). The results were similar to the main findings from the multivariable adjusted model above (i.e., HR 1.33 [95% CI 1.04–1.70] for quartile 4 vs. quartile 1 after adjustments in model 4).

CONCLUSIONS

The results of this observational study demonstrate that elevated levels of plasma ErbB2 are associated with a significantly higher risk of developing diabetes. This association remained significant after adjusting for potential confounding factors.

Circulating levels of ErbB2 have been shown to represent ErbB2/HER2 signaling activity (6). Much of the research on ErbB2 signaling has focused on its role in carcinogenesis. However, recent studies showing an association between ErbB2 and hyperglycemia and insulin resistance suggest its role beyond oncogenesis (24,30). A study conducted by Fernández-Real et al. (24) assessed the relationship of circulating ErbB2 levels with insulin resistance. The results indicated that serum ErbB2 levels were significantly associated with insulin resistance and decreased after weight loss in obese subjects, indicating that ErbB2 might have a role in the pathophysiology of diabetes (24). Additionally, a cross-sectional study by Memon et al. (30) showed a significant association between ErbB2 and hyperglycemia and insulin resistance. The cross-sectional results of our study show a similar association.

ErbB2 is a known marker for cancer, and its importance is well established in monitoring the prognosis and in the therapeutic management of breast cancers. The potential mechanism explaining the association between ErbB2 and diabetes is not clear. However, plausible biological mechanisms can be suggested to account for this association. One possible speculation regarding the link between ErbB2 and diabetes can be explained through fatty acid synthase (FASN) activity. Overexpression of ErbB2 has been correlated with increased expression of FASN (31). Some studies have explored the role of FASN as a possible surrogate marker for diabetes (32), and it has been associated with insulin resistance and type 2 diabetes (33). Therefore, we can speculate that FASN mediates the effects of ErbB2 in the development of diabetes, or perhaps a synergistic effect occurs. ErbB2 has been closely related to a number of enzymes associated with

Table 3—Incidence of diabetes in relation to ErbB2 quartiles (n = 4,220)

	Quartile 1	Quartile 2	Quartile 3	Quartile 4	P for trend	HR per 1 SD
ErbB2						
Participants, n	1,055	1,055	1,055	1,055		
Incidence of diabetes, n (per 1,000 person-years)	111 (4.91)	127 (5.92)	145 (6.86)	232 (11.57)		
HR						
Model 1	1	1.22 (0.94–1.57)	1.41 (1.10–1.81)**	2.41 (1.92–3.0)***	<0.001	1.52 (1.39–1.66)***
Model 2	1	1.08 (0.84–1.40)	1.19 (0.93–1.53)	1.72 (1.36–2.18)***	<0.001	1.25 (1.14–1.37)***
Model 3	1	1.09 (0.85–1.41)	1.19 (0.93–1.53)	1.73 (1.37–2.19)***	<0.001	1.26 (1.15–1.38)***
Model 4	1	0.93 (0.72–1.20)	0.99 (0.77–1.27)	1.31 (1.03–1.66)*	0.006	1.15 (1.05–1.25)**

Data are HR (95% CI) unless otherwise specified. Model 1 was the crude model. Model 2 was adjusted for age, sex, waist circumference, smoking habits, LDL cholesterol, systolic blood pressure, and antihypertensive drug medication. Model 3 was adjusted for model 2 variables and CRP. Model 4 was adjusted for model 3 variables and fasting glucose. HR per 1 SD, HR per 1 SD of the log2-transformed value. *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

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lipid metabolism, such as FASN, phosphatidylinositol 3 kinase, and mammalian target of rapamycin (21). Moreover, proteomic studies conducted by Zhang et al. (34) showed that ErbB2-positive cancers were associated with enhanced glycolysis, either directly or indirectly, through increased activation of various metabolic stress-responsive processes. ErbB2 has also been suggested to play a role in preadipocyte differentiation. Vazquez-Martin et al. (7) demonstrated that ErbB2 affects the process of adipocyte differentiation and therefore has a role in obesity. This close relationship with adiposity is intriguing with regard to the possible role of ErbB2 in diabetes development. Notably, both cancer and diabetes have been associated with inflammation and oxidative stress (35). It is reasonable to believe that having a common root cause might enable mechanisms to cross over, connecting both conditions.

The literature documents the antineoplastic effects of antidiabetes drug use in patients with diabetes. The drug metformin has been shown to downregulate *HER2* expression (36). On the other hand, it has also been shown that tyrosine kinase inhibitors, which typically are used as anticancer drugs, also affect glucose metabolism. Tyrosine kinase inhibitors have been suggested as a potential treatment for diabetes (37,38). The present prospective study links ErbB2 with diabetes and adds to the current knowledge, showing that further exploring this association may have value. What remains to be elucidated is the pathophysiological mechanism underlying this link. The clinical significance of ErbB2 is well established in oncology for its use in monitoring the progression of cancer. Consequently, taking into account these findings and the possible role of tyrosine kinase inhibitors in diabetes treatment (37,38), ErbB2 may have a possible additional role as a novel target for diabetes-specific therapies. Further clinical studies are warranted in order to explore this prospect.

The strengths of the study include the long follow-up period and its prospective design, which previous studies have lacked. The diabetes cases were identified through linkages with validated local and national registers. However, some limitations need to be considered. One is the lack of serial measurements of ErbB2 and risk factors over time. We adjusted for potential confounders, and the results were largely independent of these variables. Residual confounding is, however, always a potential limitation and cannot be ruled out entirely. Another aspect to consider is that the measurement for ErbB2 was expressed as arbitrary units and therefore cannot be compared with absolute values. However, the association shows that higher levels of ErbB2 are associated with an increased risk of diabetes.

In this study, complete information to distinguish diabetes types was not available. Because the study population included older adults, and because prevalent cases of diabetes were excluded, it is very likely that incident cases were type 2 diabetes. About 50% of cases were classified as type 2, but the subtype was missing for the remaining cases. However, we performed a sensitivity analysis that excluded individuals with known type 1 diabetes or LADA and those who were treated with insulin within 3 years after diabetes had been diagnosed, and the results were essentially unchanged. Another potential limitation is that we measured ErbB2 once at baseline, and blood samples were frozen and stored at −80°C. The long-term stability of ErbB2 in these samples is not known. Any minor loss of protein over time would, in our opinion, not affect the positive association identified from the results. Rather, the true effect in the study population would be underestimated.

In conclusion, in addition to breast cancer, circulating ErbB2 levels are positively associated with an increased risk of diabetes. Our results are interesting and worth exploring regarding the potential role of ErbB2 in the development of diabetes and in novel therapeutic approaches.

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