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The association between different night shiftwork factors and breast cancer: a case–control study

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Background: Research on the possible association between shiftwork and breast cancer is complicated because there are many different shiftwork factors, which might be involved including: light at night, phase shift, sleep disruption and changes in lifestyle factors while on shiftwork (diet, physical activity, alcohol intake and low sun exposure).

Methods: We conducted a population-based case–control study in Western Australia from 2009 to 2011 with 1205 incident breast cancer cases and 1789 frequency age-matched controls. A self-administered questionnaire was used to collect demographic, reproductive, and lifestyle factors and lifetime occupational history and a telephone interview was used to obtain further details about the shiftwork factors listed above.

Results: A small increase in risk was suggested for those ever doing the graveyard shift (work between midnight and 0500 hours) and breast cancer (odds ratio (OR) = 1.16, 95% confidence interval (CI) = 0.97–1.39). For phase shift, we found a 22% increase in breast cancer risk (OR = 1.22, 95% CI = 1.01–1.47) with a statistically significant dose–response relationship (P = 0.04). For the other shiftwork factors, risks were marginally elevated and not statistically significant.

Conclusion: We found some evidence that some of the factors involved in shiftwork may be associated with breast cancer but the ORs were low and there were inconsistencies in duration and dose–response relationships.

The International Agency for Research in Cancer concluded in 2007 that 'shiftwork that involves circadian disruption is probably carcinogenic to humans' (Group 2A) (IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2010). While studies of shiftwork and breast cancer to date have not shown consistent results, meta-analyses have found a 40–50% increase in risk of breast cancer for women who work at night (Megdal *et al*, 2005; Erren *et al*, 2008). Two meta-analyses published this year have included 15 and 16 studies and concluded that the evidence is insufficient to determine whether shiftwork is associated with breast cancer risk (Ijaz *et al*, in press; Kamdar *et al*, 2013). The studies included have used a range of metrics to measure the concept of shiftwork, such as ever doing night work, total number of years doing night work, number of shifts in a row and total number of night shifts worked (Tynes *et al*, 1996; Davis *et al*, 2001; Hansen, 2001; Schernhammer *et al*, 2001, 2006; Lie *et al*, 2006, 2011;

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O'Leary *et al,* 2006; Pesch *et al,* 2010; Pronk *et al,* 2010; Hansen and Lassen, 2012; Menegaux *et al,* 2012; Grundy *et al,* in press; Knutsson *et al,* 2013). In addition, different definitions of what constitutes night work have been used, such as different hours of start and finish and whether the shifts are rotating or not. Not all of these studies have specified the biological basis for their exposure variables. Frequent jet lag may have similar effects to shiftwork as demonstrated in studies of flight attendants (Erren *et al,* 2008). The differences in exposure definitions may have contributed to the debate regarding whether or not shiftwork is associated with breast cancer.

As a result of the complexity of shiftwork rosters and workplace characteristics across different industries and companies, a more useful approach is to measure exposure to particular shiftwork factors that are plausibly associated with the carcinogenic process. We thought it was appropriate to consider the biological basis by which various shiftwork characteristics might lead to breast cancer (Fritschi *et al,* 2011). First, exposure to light at night during shiftwork may suppress melatonin. Melatonin is a hormone integral to the sleep–wake cycle and evidence suggests that it may also have anticancer activity (IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2010). Second, multiple nights in a row may result in phase shift, in which the central sleep–wake cycle becomes adjusted to being awake at night. In these circumstances, the slower-adapting peripheral rhythms, including cell division, may lag behind. Third, night shiftworkers may have shorter duration or poorer quality sleep. Finally, shiftwork may induce changes in lifestyle factors such as, poorer quality diets, less physical activity, higher alcohol intake and less time outdoors (which may be associated with lower sun exposure and lower vitamin D levels).

In addition to the above complexity involved in assessing exposure to shiftwork, the reaction to shiftwork differs according to innate characteristics of the individual shiftworker. In regulating the sleep–wake cycles in humans, the circadian rhythm has been described in terms of three characteristics: phase, amplitude and stability (Horne and Ostberg, 1977; Monk and Folkard, 1985). The phase of the rhythm is commonly described as an innate preference for 'morningness' or 'eveningness'. Morning types prefer to go to bed earlier, wake up earlier and be active in the morning, whereas evening types have a preference for going to bed later, waking up later and being active in the evening. There is some evidence that morning types are more drowsy during night shifts (Smith *et al,* 2005) but this is not consistent (Sack *et al,* 2007). The amplitude of the rhythm can be measured as languidness or vigorousness (di Milia *et al,* 2005). Vigorousness represents the ability to more easily overcome tiredness following reduced sleep, while more languid types experience lethargy and persistent drowsiness. The stability of the rhythm is reported as the flexibility or rigidity of sleeping habits (di Milia *et al,* 2005). People with rigid circadian rhythms are reported to be less able to go to bed early, sleep-in or sleep at unusual times. There is some evidence to suggest that more languid types and more flexible types show better adjustment to shiftwork (Saksvik *et al,* 2011).

The aim of this population-based case–control study was to investigate the associations between the risk of breast cancer and each of the *a priori* biologically based shiftwork factors. In addition, we investigated whether this relationship was modified by circadian preference (Erren, 2010).

MATERIALS AND METHODS

Study population. Women aged between 18 and 80 years who had a first incident invasive breast cancer diagnosed between May 2009 and January 2011 were recruited for the Breast Cancer Employment and Environment Study (BCEES) (Girschik *et al,* 2012). We identified cases from the population-based Western Australian (WA) Cancer Registry, based on mandatory reporting of invasive cancer by pathology laboratories and other clinical sites. We excluded cases if their diagnosis was ductal carcinoma *in situ* or was not primary breast cancer, they previously had breast cancer, or their diagnosis date was >213 days before the cancer registry report.

During the same time period, we randomly selected control women from the WA electoral roll, and frequency age-matched them to the expected distribution of cases. Enrollment to vote is compulsory for adult Australian citizens. Women with a previous diagnosis of invasive breast cancer were excluded. Further ineligibility criteria for both case and control participants included: incorrect address, deceased, too unwell to participate, inadequate English and not resident in WA.

Approval for the study was obtained from the Human Research Ethics Committee of The University of WA and the WA Department of Health. Informed consent was obtained for all participants.

A total of 2222 cases of breast cancer were reported to the WA Cancer Registry during the recruitment period, of whom 138 were ineligible (77 with delayed registration, 14 with incorrect address, 14 with inadequate English, 12 with other serious illnesses, 8 deceased, 7 not an incident breast cancer and 6 not WA residents). Of the 2084 eligible cases invited to participate, 1205 consented (57.8%), 334 refused and 545 did not respond. From the electoral roll, 4608 women's names were extracted as controls and 252 were ineligible (155 incorrect address, 23 with previous breast cancer, 18 with serious other illness, 37 with inadequate English comprehension, 4 non-residents, 2 males, 2 deceased, and 11 for whom the reason was not recorded). Of eligible controls, 1789 (41.1%) consented to participate, 939 refused and 1628 failed to respond. Three cases and four controls did not complete the occupational section of the questionnaire.

Data collection. Data collection was completed in two stages: a questionnaire and a follow-up telephone interview. Cases and controls were sent an invitation letter, consent forms and questionnaire. The letter for the cases was sent from the WA Cancer Registry and, when the woman consented, her details were released to the study team. Controls were sent a letter by the study team. Fourteen days after initial contact, all women who had not yet responded were telephoned if a phone number was available. A reminder letter was sent 28 days after the initial contact to any non-respondents.

Women were mailed a questionnaire regarding demographic characteristics, reproductive history and lifestyle factors (e.g., alcohol intake, smoking, physical activity and sleep). They completed the Horne–Ostberg questionnaire (Horne and Ostberg, 1977), which has been shown to correlate with objective circadian phase measurements (Sack *et al,* 2007) and the Circadian Type Inventory, which measures circadian amplitude and stability (di Milia *et al,* 2005). We used the postcode of residence to assign an index of relative socioeconomic advantage and disadvantage (socioeconomic index for areas; Australian Bureau of Statistics, 2008) and an index of remoteness of residence from four classes of service centre (ARIA) (Department of Health and Aged Care and the University of Adelaide, 1999). Women were also asked their family history of breast cancer, and were classified as having a clear high risk (a first-degree female relative diagnosed with breast cancer before the age of 50, two or more first-degree female relatives diagnosed at any age, or two or more first-degree or second-degree female relatives on the same side of the family diagnosed), some family history (any first-degree or second-degree female relative diagnosed with breast cancer at any age), or no family history.

Participants also provided information on each job they had held for at least 6 months including age started, duration in years, job title, main duties, employer, industry, hours per week and weeks per year worked, and whether the job involved night work, shiftwork, work at unusual hours or flying for work purposes more than once a month.

We used structured telephone interviews by trained interviewers, blinded to disease status, to collect more detailed occupational information. Answers to the questions were recorded in OccIDEAS, an online application, which manages the interview and the process of assessing occupational exposure (Fritschi *et al,* 2009).

An interview was allocated to any job for which the women identified that they worked shifts, or for jobs likely to involve shiftwork (e.g., nurse, driver, cleaner, chef and laboratory worker). The questions included: type of roster (regular patterns, no pattern, business hours, plus on call); whether they worked for any number of hours between midnight and 0500 hours (graveyard shift); and whether they worked a shift that started between 0500 and 0700 hours. For jobs with more than one graveyard shift in a row, we asked for further details based on our *a priori* mechanistic framework (Fritschi *et al,* 2011). We also had specific questionnaires for flight attendants and people who flew frequently for work.

Exposure assessment. On the basis of the answers in the interviews in OccIDEAS, the program provided automatic assessments of the probability of exposure to the hypothesised factors (probable, possible or no exposure) based on *a priori* algorithms developed using previous literature and expert opinion. For those jobs assessed as having probable exposure, algorithms were also used to assign levels of exposure (low, medium and high).

Light at night was assessed by asking women whether they could read easily at night at work (high exposure), or could see but not well enough to read at work (medium exposure). Those women who did not fit either of these definitions, but had enough light to read in their bedroom when sleeping during the day were assigned low exposure.

For phase shift, we wished to identify those women whose central cycle had adapted to night shift but whose peripheral rhythms may not have. The central cycle is quicker to adjust when doing forward rotation rosters in which night shifts follow day shifts and then days off, than backward rotation in which day shifts follow night shifts (Haus and Smolensky, 2013). If the shift schedule had no pattern, or if there were >2 days off between finishing day and starting night shift, we conservatively assumed backward rotation, which required the woman to work a greater number of night shifts before being categorised as having high level exposure than a forward rotation. Exposure was classified as high if the job involved >4 nights forward rotation or >6 nights backward rotation, medium with 3–4 nights forward or 4–6 nights backward rotation, and low with 3 nights backward rotation. If night shift was done for more than a 4-week block, we downgraded the phase shift by one level on the assumption that peripheral rhythms would synchronise with central rhythms over this time.

Sleep disturbance was classified as high if hours of sleep were <6 or >9 h per night, sleep quality was rated quite bad or extremely bad, and they frequently or always had difficulty falling asleep or staying asleep. Medium and low exposures were assigned to those with the same durations of sleep as high level disturbance but with less frequent difficulty in falling or staying asleep. We excluded 17 women from the sleep disruption analysis who had symptoms of sleep disruption but slept between 6 and 9 h a night.

Each lifestyle factor was assessed using one or two questions regarding behaviour while on night shift. Diet was self-assessed on a four-point scale from very healthy (defined as: 'lots of vegetables and wholegrain cereals, fruits and some proteins – fish, meat legumes') to very unhealthy ('mostly fatty and sweet foods'). We asked how often the shiftworker used alcohol to fall asleep (range from almost always to never/almost never). Physical activity was assessed using two questions regarding how many times a week shiftworkers did 20 or more minutes of vigorous-intensity, or 30 min or more of moderate-intensity physical activity. For a proxy variable or vitamin D, we asked how much time was spent outside between two consecutive night shifts (options varying from <10 min to >1 h).

To limit respondent burden, where a woman had multiple jobs, we chose up to five of her jobs for the interview. These were usually the longest held, or most different from other jobs. This process resulted in 490 jobs for which we did not have phone interviews (2.5% of total). Where possible ($n=455$ jobs), we used the assessments from the woman's most similar job. For the 35 jobs for which the woman had no similar job (0.2% of jobs), we used a standard assessment based on the mode exposures in the other questionnaires (medium exposure to light at night and low exposure to phase shift, poor diet, and lack of physical activity).

Assessments for jobs were combined for each person by factor. There were a small number of jobs (<10) for which the woman's schedule was so irregular it was not possible for her to answer all the questions we asked. In these cases, we assigned possible exposure to the factors related to the difficult questions, and, if that was the only job in which they were exposed to that factor, we omitted them from the analysis for that factor. The following metrics were used in analyses: ever/never probably exposed; highest level ever probably exposed; and duration of probable exposure (in years) at medium or high level. For the lifestyle factors, we only present ever/never exposed, because the exposure assessment was based on very few questions and we had limited data on these factors when not doing night shift.

Statistical analysis. We undertook unconditional logistic regression for each of the factors separately, adjusting for 5-year age group, to calculate odds ratios (ORs) and 95% confidence intervals (CIs). The reference group was women without exposure to that factor whether or not they had done shiftwork. We repeated the analysis after excluding women who had done the graveyard shift but never been exposed to any of the other shiftwork variables ($n=71$). The known risk factors for breast cancer were associated with breast cancer in the expected direction (Table 1). We tested the effect of adding each of these 10 categorical risk factors to the model for each of the four main exposure variables (graveyard shift, light at night, phase shift and sleep disruption) and three lifestyle factors (poor diet, lack of physical activity and little time outdoors) using the chest command in Stata 12 (StataCorp, College Station, TX, USA). Adding any of these risk factors to the models containing age group and the exposure variable of interest did not change the OR for the exposure variable by $>2\%$, so to keep models parsimonious they were not included in the final models.

We stratified the analyses for morningness/eveningness (Horne and Ostberg, 1977), flexibility/rigidity and languidness/vigorousness of circadian rhythm (di Milia *et al,* 2005) and menopausal status at time of recruitment.

In order to examine latency, we repeated the analysis indicating whether exposure occurred in the windows of time >30 years, >20 and $\leqslant 30$ years, >10 and $\leqslant 20$ years, and $\leqslant 10$ years before recruitment compared with those who were unexposed during that window of time. Subjects could contribute to more than one of these exposure variables.

We also repeated analyses for exposure before the first birth (restricting analysis to women with at least one child).

As the different shiftwork metrics were moderately to highly correlated (ranging from 0.16 to 0.91) we also assessed all metrics simultaneously by two novel methods. First, by using a Bayesian mixture framework developed for the analysis of correlated

Table 1. Demographic and reproductive characteristics of breast cancer cases and controls, 2009–2011

	% Controls (**n** = 1785)	% Cases (**n** = 1202)
Age group (years)		
24–44	10.0	14.0
45–49	11.4	14.6
50–54	13.3	12.7
55–59	17.1	15.3
60–64	18.0	16.0
65–69	15.2	12.2
70–80	15.0	15.3
Status at recruitment		
Postmenopausal	76.5	69.6
Premenopausal	23.5	30.4
Socioeconomic score[a]		
Advantaged	5.1	4.5
2	13.7	12.7
3	20.8	19.5
4	20.1	20.5
Disadvantaged	40.2	42.9
Remoteness score		
Highly accessible	84.6	85.7
Less accessible/remote	15.4	14.3
Education		
Junior school	36.0	36.1
Senior school	22.6	20.6
Trade/apprenticeship	24.4	21.5
University	17.0	21.8
Country of birth		
Australia/New Zealand	66.6	63.7
United Kingdom/ Ireland	21.6	21.7
Other Europe	4.7	5.4
Asia	3.7	5.3
Other	3.4	3.8
Family history of breast cancer		
None	71.6	60.6
Some family history	20.9	25.0
Clear high risk	7.3	14.1
Unknown or missing	0.2	0.3
No. of children		
0	10.5	13.1
1	7.7	9.6
2	38.9	38.2
3	27.2	25.1
4 +	15.7	14.1
Breastfeeding[b]		
No children	10.5	13.1
No breastfeeding	8.6	10.5
Breastfed ⩽ 12 months	35.6	32.8
Breastfed > 12 months	43.2	42.2
Alcohol intake[c,d]		
None	15.9	16.8
⩽3 drinks per week	36.3	34.6
4–9 drinks per week	21.2	22.3
10 + drinks per week	26.2	26.1
Physical activity[d,e]		
0–37	24.7	24.7
38–58	25.2	24.7
59–85	25.1	26.2
86 +	25.0	24.4
BMI in 30s[f]		
Underweight	8.8	8.0
Healthy	70.8	72.5
Overweight	15.2	13.8
Obese	5.2	5.7
Circadian type[g]		
Evening type	20.0	20.6
Neutral type	51.8	49.4
Morning type	27.3	28.6
Circadian rhythm[h]		
Vigorous	19.0	19.8
Neutral	48.4	50.7
Languid	31.9	28.6
Circadian flexibility[i]		
Rigid	21.6	22.8
Neutral	52.9	51.1
Flexible	24.9	25.1

Abbreviations: BMI = body mass index; MET = metabolic equivalent of task. Missing values: [a]1; [b]54; [c]9; [f]354; [g]32; [h]23; [i]24 were dropped from the analysis.
[d]These data relate to alcohol and physical activity overall, rather than related to shiftwork.
[e]Quartiles of average MET-hours per week in all activity over the lifetime, 59 missing values.

exposure variables in multiple logistic regression (de Vocht *et al*, 2012). Second, using an elastic net analyses (i.e., GLMNET analysis), which combines two penalised regression models (Lasso and Ridge). In the elastic net model, the constraint is defined as a balance between L^1 (Lasso) and L^2 (Ridge) norms of the regression coefficients, which is specified by λ. When $\lambda = 0$, it exactly corresponds to the Lasso regression (*a priori* expectation is that only one of the correlated shiftwork factors is responsible for the association) and if $\lambda = 1$, to the Ridge regression model (*a priori* expectation is that all shiftwork factors contribute to the association; Friedman *et al*, 2010).

At the 5% level of significance, we had 80% power to detect an ORs of 1.5 if the prevalence of medium-to-high exposure to shiftwork was at least 4%.

RESULTS

Cases who refused to participate were older than participants, while those who did not respond were younger and were more likely than responders to live in very remote areas (4% *vs* 2%). There were no differences in socioeconomic status between the three groups of cases. Controls who refused were older than those who participated, while those who did not respond were younger but there were no differences in residential remoteness or socioeconomic status between the three groups.

Despite the age-frequency matching, differential response fractions by age group resulted in the controls being slightly older than the cases, and a greater proportion of controls were post-menopausal (Table 1). Cases tended to have higher education than controls but had similar distributions for SEIFA and ARIA. The expected patterns of known risk factors were seen in our participants with cases being more likely to have a family history of breast cancer, more likely to be nulliparous and less likely to have breastfed their children.

For 2630 (13.7%) of the total of 19 214 jobs, participants indicated that they did 'night work, shiftwork or work at unusual hours'. Of these jobs: 954 involved regular patterns of shift and in 440 (46.7%) the woman worked graveyard shifts; 1513 jobs involved varied rosters of which 1063 (70.1%) included graveyard shifts; and 163 involved regular hours plus on call of which 95 (59.0%) included graveyard shifts. The majority of shiftwork jobs (57.2%) were health-related, primarily nursing. Other shiftwork jobs included workers in entertainment and hospitality (such as bars, restaurants, accommodation, sports venues and media – 15.2% of shiftwork jobs), cleaners (6.7%), production workers (4.6%), drivers (2.6%), telephonists/call centre operators/data entry workers (2.6%) and laboratory workers (1.8%).

There were a total of 670 women who had ever done a graveyard shift and their age-adjusted risk of breast cancer was 1.16 (95% CI = 0.98–1.39) but there was no duration–response relationship.

When adjusted for age group, exposures to light at night, phase shift and sleep disruption all were associated with slightly increased risks of breast cancer (Table 2), but the results reached statistical significance only for phase shift (OR 1.22, 95% CI = 1.01–1.47).

There was a statistically significant dose–response relationship for phase shift ($P = 0.04$) but no duration–response association. There were no statistically significant relationships between breast cancer and dose or duration of exposure to light at night or sleep disruption.

Associations between the reported lifestyle factors during night shift and breast cancer were as follows: poor diet OR = 1.21 95% CI = 1.00–1.46; alcohol to help sleep OR = 1.64 95% CI = 0.64–4.20; lack of physical activity OR = 1.20 95% CI = 0.98–1.47; and little time outdoors OR = 1.21 95% CI = 0.94–1.54.

No substantial differences were seen when we omitted women whose exposure occurred solely as a result of work as a flight attendant ($n = 10$), nor women who had done the graveyard shift but never been exposed to any of the other shiftwork variables ($n = 71$; data not shown).

The data were further analysed using the multivariate Bayesian mixture framework including all hypothesised mechanisms and additional adjustment for age. Assuming no prior knowledge on the causal mechanism the prior probability for each hypothesised mechanism to be associated with breast cancer with set at 50%, the model results indicated that no single variable was associated with breast cancer. Posterior ORs range 1.00 to 1.01 (i.e., the prior distribution of no effect) and 95% credible intervals all included unity. Posterior probabilities that a specific variable had an effect on breast cancer ranged from 24% to 41%, with the highest posterior probabilities observed for 'alcohol to help sleep' (41%), 'phase shift' (40%) and 'poor diet' (40%).

The elastic net analyses clearly favoured the Ridge penalty ($\lambda = 0.1$), suggesting that the shiftwork variables all contributed (nonsignificantly) to the risk of breast cancer (data not shown).

Stratification by morningness/eveningness showed a tendency for the associations between the exposures and breast cancer to be higher among both neutral and morning preference women than among evening preference women (Table 3). The only statistically significant associations were seen among neutral preference type women for graveyard shift (OR = 1.34, 95% CI = 1.04–1.73) and for phase shift (OR = 1.34 95% CI = 1.02–1.77). However, none of the interaction terms were statistically significant.

Table 2. Age group-adjusted associations between risk of breast cancer and exposure to graveyard shift, light at night, phase shift and sleep disruption

Exposure	Controls	Cases	OR (95% CI)
Graveyard shift			
Never	1404	914	Reference
Ever	381	288	1.16 (0.97–1.38)
Duration of exposure			
<10 years	199	164	1.25 (1.00–1.56)
10–<20 years	98	71	1.09 (0.79–1.50)
20+ years	84	53	1.02 (0.71–1.45)
Light at night			
Never	1447	947	Reference
Ever	335	253	1.15 (0.96–1.38)
Highest dose of exposure			
Low	3	0	*
Medium	160	110	1.06 (0.82–1.37)
High	172	143	1.25 (0.98–1.59)
Duration of exposure at medium and/or high levels			
<10 years	186	153	1.25 (0.99–1.57)
10–<20 years	80	65	1.21 (0.86–1.70)
20+ years	66	35	0.84 (0.55–1.28)
Phase shift			
None	1476	959	Reference
Ever	309	242	1.22 (1.01–1.47)
Highest dose of exposure			
Low	51	36	1.09 (0.70–1.68)
Medium	1.21177	140	1.24 (0.97–1.57)
High	81	66	1.25 (0.90–1.75)
Duration of exposure at medium and/or high levels			
<10 years	160	140	1.35 (1.06–1.72)
10–<20 years	58	42	1.12 (0.74–1.68)
20+ years	40	24	0.96 (0.58–1.61)
Sleep disruption			
None	1618	1067	Reference
Ever	158	127	1.21 (0.95–1.55)
Highest dose of exposure			
Low	49	41	1.27 (0.83–1.94)
Medium	40	34	1.25 (0.79–2.00)
High	69	52	1.14 (0.79–1.66)
Duration of exposure at medium and/or high levels			
<10 years	61	55	1.32 (0.91–1.92)
10–<20 years	27	16	0.87 (0.46–1.63)
20+ years	21	15	1.15 (0.59–2.25)

Abbreviations: CI = confidence interval; OR = odds ratio.

There were no statistically significant associations between the exposures and breast cancer after stratification by menopausal status, or circadian amplitude or stability (Table 3).

Our latency analyses showed a tendency for exposure to graveyard shift or sleep disruption, which occurred >30 years ago to have a slightly stronger effect than more recent exposure although no results were statistically significant (Table 4).

Table 3. Associations between ever exposed to light at night, phase shift and sleep disruption during night shift stratified by different variables (all age group-adjusted)

Stratifying variables	*N*[a] total	Graveyard OR (95% CI)	*N*[a]	Light at night OR (95% CI)	*N*[a]	Phase shift OR (95% CI)	*N*[a]	Sleep disruption OR (95% CI)	*N*[a]
Circadian preference									
Evening type	248/357	0.95 (0.66–1.38)	66/97	0.96 (0.65–1.42)	57/83	1.02 (0.68–1.52)	53/75	1.15 (0.68–1.92)	30/39
Neutral type	594/924	1.34 (1.04–1.73)	136/170	1.25 (0.96–1.64)	119/155	1.34 (1.02–1.77)	112/139	1.22 (0.84–1.77)	54/71
Morning type	344/488	1.12 (0.81–1.55)	86/111	1.20 (0.85–1.68)	77/95	1.23 (0.87–1.72)	77/93	1.30 (0.84–2.04)	43/46
Circadian amplitude									
Vigorous	238/340	1.20 (0.79–1.82)	52/65	1.06 (0.68–1.64)	43/58	1.12 (0.72–1.74)	42/56	1.03 (0.55–1.94)	19/26
Neutral	610/863	1.12 (0.88–1.43)	155/200	1.21 (0.94–1.55)	144/175	1.25 (0.97–1.61)	139/165	1.26 (0.90–1.77)	72/82
Languid	344/569	1.22 (0.88–1.70)	78/114	1.09 (0.76–1.55)	63/100	1.17 (0.81–1.69)	58/87	1.24 (0.78–1.97)	35/49
Circadian stability									
Rigid	274/385	1.16 (0.77–1.77)	47/61	1.10 (0.70–1.71)	40/53	1.10 (0.70–1.73)	39/52	1.48 (0.85–2.59)	28/27
Neutral	614/944	1.28 (1.00–1.65)	138/173	1.25 (0.96–1.63)	122/155	1.26 (0.96–1.66)	112/143	1.16 (0.80–1.66)	56/76
Flexible	302/444	1.00 (0.73–1.37)	101/146	1.05 (0.76–1.45)	90/126	1.23 (0.89–1.71)	90/113	1.00 (0.71–1.70)	42/55
Menopausal status at time of recruitment									
Postmenopausal	366/419	1.18 (0.96–1.45)	196/285	1.17 (0.94–1.45)	170/247	1.21 (0.97–1.51)	168/236	1.28 (0.94–1.72)	84/112
Premenopausal	836/1366	1.13 (0.81–1.57)	92/96	1.10 (0.78–1.55)	83/88	1.22 (0.85–1.74)	74/73	1.08 (0.69–1.68)	43/46

Abbreviations: CI = confidence interval; OR = odds ratio.
[a]Number of subjects exposed to this agent – cases–controls.

Table 4. Risk of breast cancer associated with exposure to three shiftwork factors when exposure occurred at different windows of time relative to interview

Window of exposure	Exposure during that window	Graveyard shift OR (95% CI)	Light at night OR (95% CI)	Phase shift OR (95% CI)	Sleep disruption OR (95% CI)
⩽10 years ago	No	1.0	1.0	1.0	1.0
	Yes	1.02 (0.73–1.43)	1.25 (0.86–1.80)	1.23 (0.83–1.83)	1.10 (0.68–1.79)
>10 and ⩽20 years ago	No	1.0	1.0	1.0	1.0
	Yes	1.08 (0.77–1.51)	0.89 (0.61–1.30)	0.98 (0.66–1.46)	0.82 (0.49–1.37)
>20 and ⩽30 years ago	No	1.0	1.0	1.0	1.0
	Yes	0.83 (0.60–1.14)	0.87 (0.62–1.21)	0.86 (0.61–1.22)	1.07 (0.67–1.72)
>30 years ago	No	1.0	1.0	1.0	1.0
	Yes	1.21 (0.94–1.57)	1.22 (0.93–1.59)	1.19 (0.91–1.55)	1.38 (0.91–2.09)

Abbreviations: CI = confidence interval; OR = odds ratio. Exposure in each time window was compared with the reference category of never exposed at that time. All analyses are adjusted for age group and all other time window variables in the table.

We restricted the analysis to women who had one or more children, and, after adjusting for age group, there were no associations between breast cancer and exposure before the birth of the first child to the graveyard shift (OR = 0.98, 95% CI = 0.79–1.22), light at night (OR = 1.00, 95% CI = 0.79–1.25), phase shift (OR = 1.02, 95% CI = 0.82–1.29) or sleep disruption (OR = 1.02, 95% CI = 0.73–1.43).

DISCUSSION

This study of shiftwork and breast cancer investigated specific *a priori* hypotheses based on biologically plausible mechanisms (Fritschi *et al*, 2011). We found a statistically significant association with the factor of phase shift with a dose–response, but no duration–response relationship. Risks were slightly above null for the other factors but they were not statistically significant.

For comparison purposes, we analysed similar metrics to previous studies and found no association between the risk of breast cancer and having ever done graveyard shifts (OR = 1.16, 95% CI = 0.98–1.39) nor duration of work in graveyard shifts. Previous studies have found risks ranging from 0.6 to 2.9 (Davis *et al*, 2001; Hansen, 2001; O'Leary *et al*, 2006; Pesch *et al*, 2010; Pronk *et al*, 2010; Hansen and Lassen, 2012; Hansen and Stevens, 2012; Menegaux *et al*, 2012).

We found no associations between length of time exposed to any of the shiftwork-associated factors and breast cancer. Duration metrics have been used by a number of studies with the maximum category varying from ⩾4.5 to ⩾30 years (Tynes *et al*, 1996; Davis *et al*, 2001;

Hansen, 2001; Schernhammer *et al*, 2001, 2006; Lie *et al*, 2006, 2011; O'Leary *et al*, 2006; Pesch *et al*, 2010; Pronk *et al*, 2010; Hansen and Lassen, 2012; Hansen and Stevens, 2012; Menegaux *et al*, 2012). Two studies on US nurse cohorts suggested that the effect of night shift on breast cancer only occurred after >20 years of shiftwork (Schernhammer *et al*, 2001, 2006), but this has not been consistent.

There is a good theoretical basis and sufficient experimental evidence to support suppression of melatonin because of light at night as a potential cause of cancer, although under real world conditions, shiftwork has not been consistently associated with reduced melatonin levels (Schernhammer *et al*, 2004; Folkard, 2008; Grundy *et al*, 2009, 2011; Davis *et al*, 2012; Bracci *et al*, 2013). We specifically asked about the level of light in the workplace during night shifts and found little evidence of an association with breast cancer.

There was a 22% increase in risk of breast cancer in those who had ever done shiftwork in patterns that fitted our definition of inducing phase shift. There was also a tendency towards a dose–response relationship, but no duration–response relationship. However, in a multi-metric analysis using a novel Bayesian mixture model developed to deal with correlated variables this finding was not confirmed with no single factor being associated with an important increase in risk. Similarly, multi-metric elastic net analyses did not show strong evidence for a specific effect with phase shift in combination with the other parameters but provided additional evidence for nonsignificant small effects of all shiftwork factors combined.

Our definition of phase shift attempted to identify those shift patterns in which the external times forced on a woman by shiftwork were not in phase with peripheral cellular rhythms (Blask *et al*, 2011). The highest exposure was defined as occurring when the shift pattern involved more than four nights of forward rotation or more than six nights of backward rotation. These definitions were based on evidence that central cycles start to change after several days, although much of this evidence comes from studies in laboratory animals (Costa *et al*, 2010). Phase shift may result in changes in the control of cell proliferation, which may increase the risk of cancerous development (Haus and Smolensky, 2006).

Our study found little association between risk of breast cancer and lifestyle factors when doing night shift (diet, physical activity, alcohol intake and sun exposure). However, we had limited data on these exposures and had no data on women's lifestyle in relation to these factors during periods when they were not doing shiftwork.

We did not find a significant association between breast cancer risk and sleep disruption while on night shift, which was similar to our findings regarding sleep disruption and breast cancer irrespective of shiftwork (Girschik *et al*, 2012). No previous studies have specifically examined the association between breast cancer and sleep duration and quality in relation to shiftwork. Our definition of sleep disruption was quite restrictive. We excluded women with sleep disruption who had sleep durations that fell within the normal range. There are also some people who naturally do not need 6 h of sleep or need >9 h sleep but they would not have been classified as having sleep disruption unless they also had symptoms of insomnia and rated their sleep quality as poor.

The stage of life at which exposure occurs may be important for two reasons. First, there are changes in the breast architecture during the first pregnancy. We found no increase in risk estimates when we examined exposure occurring before first birth. A previous study found that doing night shifts for >4 years before first birth increased the risk of breast cancer but only in women doing <4 night shifts per week (OR 3.0, 95% CI = 1.4–6.5; Menegaux *et al*, 2012). Second, it is not clear when shiftwork may act in the development of breast cancer. In our latency analyses, we found a nonsignificant association with distant exposure (>30 years before interview), which may indicate, if anything, an inducing effect of night shift (Blask *et al*, 2011).

It has been theorised that women with a morning preference may suffer more from circadian rhythm disruption when working at night than those with an evening preference (Erren, 2010; Saksvik *et al*, 2011). We used a validated scale (Horne and Ostberg, 1977) for assessing diurnal preference and found the association between ever working the graveyard shift and breast cancer was higher in those with a neutral preference (OR = 1.34) than in those with an evening (OR = 0.95) or morning (OR = 1.12) preference, although there were no statistically significant interactions. One previous study examined diurnal preference (assessed with a single question) as a modifier of the association between breast cancer and night work and found risks raised for women with a morning preference (OR = 3.9, 95% CI = 1.6–9.5) and no association in those with neutral preference (OR = 0.7, 95% = CI 0.1–3.0; Hansen and Lassen, 2012). Our study had relatively small numbers of cases and controls in some of the subgroups, which may have resulted in limited power to assess possible effect modification, there were also multiple tests so some of the observed variation may be due to chance.

In their evaluation of the evidence regarding breast cancer and shiftwork, the International Agency for Research in Cancer highlighted that there were a limited number of professions that had been studied (IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2010). Of the 14 studies that have investigated shiftwork as a risk factor for breast cancer (Tynes *et al*, 1996; Davis *et al*, 2001; Hansen, 2001; Schernhammer *et al*, 2001, 2006; Lie *et al*, 2006, 2011; O'Leary *et al*, 2006; Pesch *et al*, 2010; Pronk *et al*, 2010; Hansen and Lassen 2012; Hansen and Stevens, 2012; Menegaux *et al*, 2012), seven have focussed on specific occupational groups, most commonly nurses and flight attendants. In contrast, our study included a wide range of shiftwork jobs with considerable variety in the rostering arrangements and characteristics of the environment in which they worked. Therefore, shiftwork is unlikely to be correlated to other occupational exposures or specific occupational groups minimising the effect of uncontrolled confounding.

We examined a large number of potential confounders (Table 1) including education, family history of breast cancer, parity, breastfeeding and physical activity. None of these affected the relationship between our shiftwork exposures and breast cancer, but we were unable to get reliable information on HRT use and there may be residual confounding from other unknown variables. One major potential confounder is body mass index (BMI). We used BMI at age 30 as preferable to measuring weight at around the time of diagnosis or treatment of breast cancer (when the disease or the treatment may have affected weight). However, the use of BMI at this age may be inaccurately recalled, and 354 women in our study were unable to provide this information resulting in a reduction in sample size.

Our study was large, but given the relatively low response fractions, there may be some selection bias. We used the methods outlined by Lash *et al* (2009) to examine what level of selection bias would hide a real effect of 1.5 for ever working the graveyard shift (Megdal *et al*, 2005). If there was no bias in control selection, the non-participating cases would need a higher prevalence of exposure (36%) than that of the responding cases (24%). If there was no bias in case selection, the non-participating controls would need a lower prevalence of exposure (15%) than that of the responding controls (21%). There were some differences in age and residential remoteness between those who participated and those who did not for cases and differences in age for controls but it is not obvious how this would result in a bias strong enough to mask a real effect of 1.5, given that shiftwork was not mentioned in study information sheets.

In conclusion, our study was designed to specifically test a comprehensive set of biologically based hypotheses regarding the association of shiftwork and breast cancer. We found a statistically significant association between risk of breast cancer and phase shift caused by night work (OR = 1.22, 95% CI = 1.01–1.47) with evidence of a dose–response but no duration–response relationship. However, there were no statistically significant associations with other factors, the association with morningness/eveningness was not in the direction as predicted by theory and multi-metric analyses provided more support for (nonsignificant) small effects related to the different shiftwork factors. The latter may indicate some small risks associated with shiftwork or some residual confounding. These results need to be repeated in other well-designed studies in order for us to determine what advice we should give to the millions of women around the world who work at night.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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