Supplementary Material

Title: Women's pregnancy life-history and Alzheimer's risk: can immunoregulation explain the link?

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	Total	Medical exclusion criteria	Missing data: Breastfeeding	Missing data: Age at menopause or menarche	Missing data: Marriages
Probands omitted	0	10	8	13	7
Resulting cohort size	133	123	115	102	95

Table S1. Cohort size

Caption: From the original recruited sample of 133 probands, 95 cases were included in analyses.

Variables tested for covariate status	Potential confound	Cumulative months pregnant: p-values	Cumulative months pregnant (median): p-values	Parity: p-values	Cumulative number of 1st trimesters	Cumulative number of 3rd trimesters
Any breastfeeding	✓	0.01 *	0.09 +	0.04 *	0.01 *	0.01 *
Cumulative breastfeeding	✓	ns	ns	ns	ns	ns
Age first birth	✓	0.001 **	0.001 **	0.02 *	0.0003 ***	0.004 **
Reproductive span	✓	0.09 +	ns	ns	0.08 +	ns
Age at menopause	✓	0.09 +	0.09 +	ns	0.06 +	ns
Age at menarche	✓	0.06 +	0.09 +	ns	ns	ns
Sum duration marriages	✓	ns	ns	ns	ns	ns
Age first married	✓	ns	ns	ns	ns	ns
Occupation	✓	0.03 *	0.06 +	0.02 *	0.02 *	0.02 *
Interaction term: Any breastfeeding	✓	ns	ns	ns	ns	ns
Interaction term: First-degree relative with dementia	✓	ns	ns	ns	ns	ns
Age at interview	×					
Age at interview (exponentiated)	×					
Over age 90 at interview	×					
Education	×					
Use of estrogen-replacement therapy	×					
Use of hormonal contraception	×					
Hysterectomy	×					
Age at hysterectomy	×					
Bilateral oophorectomy	×					

Table S2. Covariates and their contributions to model fitting.

***$p < 0.001$; **$p < 0.01$; *$p < 0.05$; +$p < 0.10$. The second column shows which variables were included in models. These assessments were made on the basis of a series of generalized linear models, linear regressions, Pearson's product-moment correlations, and Chi-squared tests. The interaction terms, breastfeeding and marital history were additionally included based on correlations reported in previous studies. For nulliparas (N=7), age at first birth was replaced with the maximum age of participants (100 years). For women without any history of marriage, age at married was replaced with age at interview. "Reproductive span" refers to months between menarche and menopause. "Occupation" refers to job held longest categorized by level of labour skill. "Education" refers to age at education cessation.

Alzheimer's age at onset determination

For those individuals with CDR-SOB scores of 0.5 or above, we estimated the age at AD onset. Typical durations of each dementia stage have been reported for a variety of diagnostic instruments including the MMSE, GDS, FAST, and CDR, but no specific durations have been reported for the CDR-SOB. We used the CDR-SOB measurement to determine age at onset because it is a more sensitive scale than Global CDR, having more stage distinctions and therefore more data points. In order to convert clinical information on timing of disease progression obtained from other instruments into timing of CDR-SOB stages, we determined how CDR-SOB stages might best line up with GDS and CDR stages (see below).

Then, we substituted the endpoints for GDS and CDR stages with corresponding CDR-SOB scores. We decided that in terms of timing progression, the starting point of each category should correspond to the starting point of the CDR-SOB phase, and therefore the ending point of each category should correspond not to the end point of the CDR-SOB phase but rather the starting point of the next CDR-SOB phase. For example, GDS=5/CDR=1 occur within years 9 to 10.5 of disease progression. The corresponding CDR-SOB range is 4.5 to 9. Therefore, we determined that the starting point of this phase occurs at CDR-SOB=4.5 and ends at CDR-SOB=9.5, indicating that the phase occurs from the start of CDR-SOB=4.5 and the next phase begins at CDR-SOB=9, or 4.5 ≤ Mild Dementia < 9.5. Such a numerical determination makes the most sense when the progression of phases is considered:

$0 \leq \text{Normal} < 0.5 \leq \text{MCI} < 3 \leq \text{Mild} < 4.5 \leq \text{Moderate} < 9.5 \leq \text{Severe} \leq 18$

The pattern exception occurs at the inclusion of 18 within the 'severe' stage, as this is the scale maximum.

a) *Normal:*

GDS=1-2
CDR=0
CDR-SOB=0
This phase represents the normal healthy adult stage that precedes cognitive decline. It is characterized by no impairments in daily living in terms of activities and hobbies, and either no memory loss or slight inconsistent forgetfulness. This phase also includes GDS=2, 'Subjective Cognitive Impairment' which is a stage characterized by the slightest complaints in memory problems such as difficulty with name recall and occasional misplacing of objects (Reisberg et al., 2010). As this description is too mild to warrant a CDR 0.5 in the "memory" category, which requires consistency in forgetfulness and only partial recollection of recent events, we have included GDS=2 as part of the CDR=0 and CDR-SOB=0 stage. This determination is consistent with Reisberg's assessment in which CDR=0 is lined up with GDS=1 and 2.

b) *Mild cognitive impairment:*

GDS=3
CDR=0.5
CDR-SOB=0.5-2.5
This is the first phase in which dementia becomes apparent. It is often referred to as Mild Cognitive Impairment (MCI), and lasts approximately 7 years in those who do go on to develop AD. However, not all people who develop MCI go on to develop AD or any form of dementia. In fact, 45% of people with MCI have no change in their condition, or even experience improved cognitive status later (Smith et al., 1996). The CDR refers to this phase as "questionable dementia" or "very mild dementia" (Morris 1993). However, the CDR has fewer stages delineated than the GDS and CDR-SOB, so we have chosen to acknowledge the important distinction between MCI and very mild dementia, rendering the phase discussed here best titled "MCI" or "questionable dementia." The CDR-SOB stages, as described by O'Bryant and colleagues (O'Bryant et al., 2008) consider this phase to be best described as 0.5-2.5 as a

subset of CDR=0.5. This range of CDR-SOB scores could be achieved by exhibiting a 0.5=questionable in five of the six CDR categories, and likely a 0 in the Personal Care category for which there is no 0.5 rating (Morris 1993). This phase would preclude individuals who exhibit a 1=mild score in any category unless other categories are rated as zero to compensate.

c) *Very mild dementia*

GDS=4
CDR=0.5
CDR-SOB=3-4
This phase represents a very mild form of dementia that is one subtle step beyond MCI. The Global CDR does not distinguish between MCI and very mild dementia, but using the CDR-SOB a distinction is apparent (O'Bryant et al., 2008). The CDR-SOB category "very mild dementia" represents slight impairment that goes beyond 0.5=questionable scores in the CDR categories, or at least a score of 1=mild in the category of Personal Care. While the Global CDR may still be assessed as 0.5=questionable, certain categories may earn scores of 1=mild. The GDS describes the phase GDS=4 as being characterized by decreased knowledge of personal and/or current events, and some minor impairment in ability to perform daily tasks such as finances or shopping (Reisberg et al., 2010). Despite the fact that the GDS stage names do not line up well with CDR or CDR-SOB, the descriptions of the stages do, as does the chart presented in Reisberg et al. 2010. Our assessment of GDS=4 corresponding to higher CDR=0.5 is consistent with Reisberg's assessment.

d) *Mild dementia*

GDS=5
CDR=1
CDR-SOB=4.5-9
This phase is characterized by moderate memory loss that interferes with daily life, and is more marked for recent events. Both the GDS=5 and CDR=1 describe how individuals as this stage are unable to function independently in the community, although they may still be involved in some community activities with the assistance of carers (Morris 1993, Reisberg et al., 2010). Both scales also describe how individuals in this phase require prompting in order to carry out personal care activities. While the title of this phase differs between CDR (mild) and GDS (moderate), the descriptions are nearly identical. This is due to the fact that GDS inserts an extra phase name between moderate and severe stages (Reisberg et al., 2010). The corresponding CDR-SOB scores for "mild" are 4.5-9, which encompasses a wide range of individual box score possibilities, all in which the majority of categories are 1=mild, as well as 3 categories 1=mild and 3 categories 2=moderate, further justifying the comparison to GDS=5. Our assessment of GDS=5 corresponding to CDR=1 is consistent with Reisberg's assessment.

e) *Moderate dementia*

GDS=6
CDR=2
CDR-SOB=9.5-15.5
This phase represents a more serious stage of memory loss and inability to function at normal levels. Both GDS and CDR scoring rules indicate that individuals at this stage experience severe memory loss in which new information is rapidly forgotten and some important long-term information may be lost. Additionally, both scales describe how individuals require assistance with basic activities of daily life and personal care, incontinence may begin, and social judgment / personality are compromised (Morris 1993, Reisberg et al., 2010). While the titles used by GDS (moderately severe) and CDR (moderate) vary slightly, the descriptions are overlapping. The CDR-SOB category of "moderate dementia" encompasses

scores ranging 9.5-15.5. Our assessment of GDS=5 corresponding to CDR=2 is consistent with Reisberg's assessment.

f) *Severe dementia*

GDS=7
CDR=3
CDR-SOB=16.0-18.0
This phase represents the most severe and final stage of the disease. Both the GDS and CDR describe severe memory loss, with only fragmentary ability to recall any personal details. The GDS scoring rules describe the physical deterioration of individuals in this phase as gradual loss of all bodily functions occurs, and the CDR scoring rules describe the deterioration of all cognitive processes such as complete inability to solve problems, complete loss of orientation, and inability to perform any tasks or activities. This phase, which precedes death, may last between 2.5-6 years. In sum, our determination of disease staging is as described in Table S3.

	None	MCI	Very Mild	Mild	Moderate	Severe
GDS	1, 2	3	4	5	6	7
CDR	0	0.5		1	2	3
CDR-SOB	0	0.5-2.5	3-4	4.5-9	9.5-15.5	16-18
Years	N/A	0-7	7-9	9-10.5	10.5-13	13-19

Table S3: Equivalent stages of dementia and corresponding typical durations.

Based on the information in Table S3, we plotted the dementia stages on the Y-axis and years since onset on the X-axis, meaning the origin was at the start of MCI. Therefore, we had 6 data points. Using the statistics program Igor Pro, we interpolated 2,000 points between the known data points, and were able to determine the year corresponding to 0.5 increments in CDR-SOB scores. The values in Table S4 were thus determined.

CDR-SOB	Interpolated years since dementia onset
0.5	0 (given)
1	1.40
1.5	2.80
2	4.20
2.5	5.60
3	7.00 (given
3.5	7.67
4	8.33
4.5	9.00 (given)
5	9.15
5.5	9.30
6	9.45
6.5	9.60
7	9.75
7.5	9.90
8	10.05
8.5	10.20
9	10.35
9.5	10.50 (given)
10	10.69
10.5	10.88

11	11.08
11.5	11.27
12	11.46
12.5	11.65
13	11.85
13.5	12.04
14	12.23
14.5	12.42
15	12.62
15.5	12.80
16	13.00 (given)
16.5 (hypothetical only)	14.50
17	16.00
17.5 (hypothetical only)	17.50
18	19.00 (given)

Table S4: Years since dementia onset as estimated from CDR-SOB score.

Data presented here are based on our assessment and published equivalencies between various diagnostic instruments including the CDR and GDS, and interpolated from typical duration of stages as reported by Reisberg (Reisberg et al., 2010). "Given" values come directly from Reisberg (Reisberg et al., 2010). "Hypothetical only" CDR-SOB scores are included only to demonstrate the method of calculation, as no combination of CDR box scores could add up to these values.

Supplementary Figures:

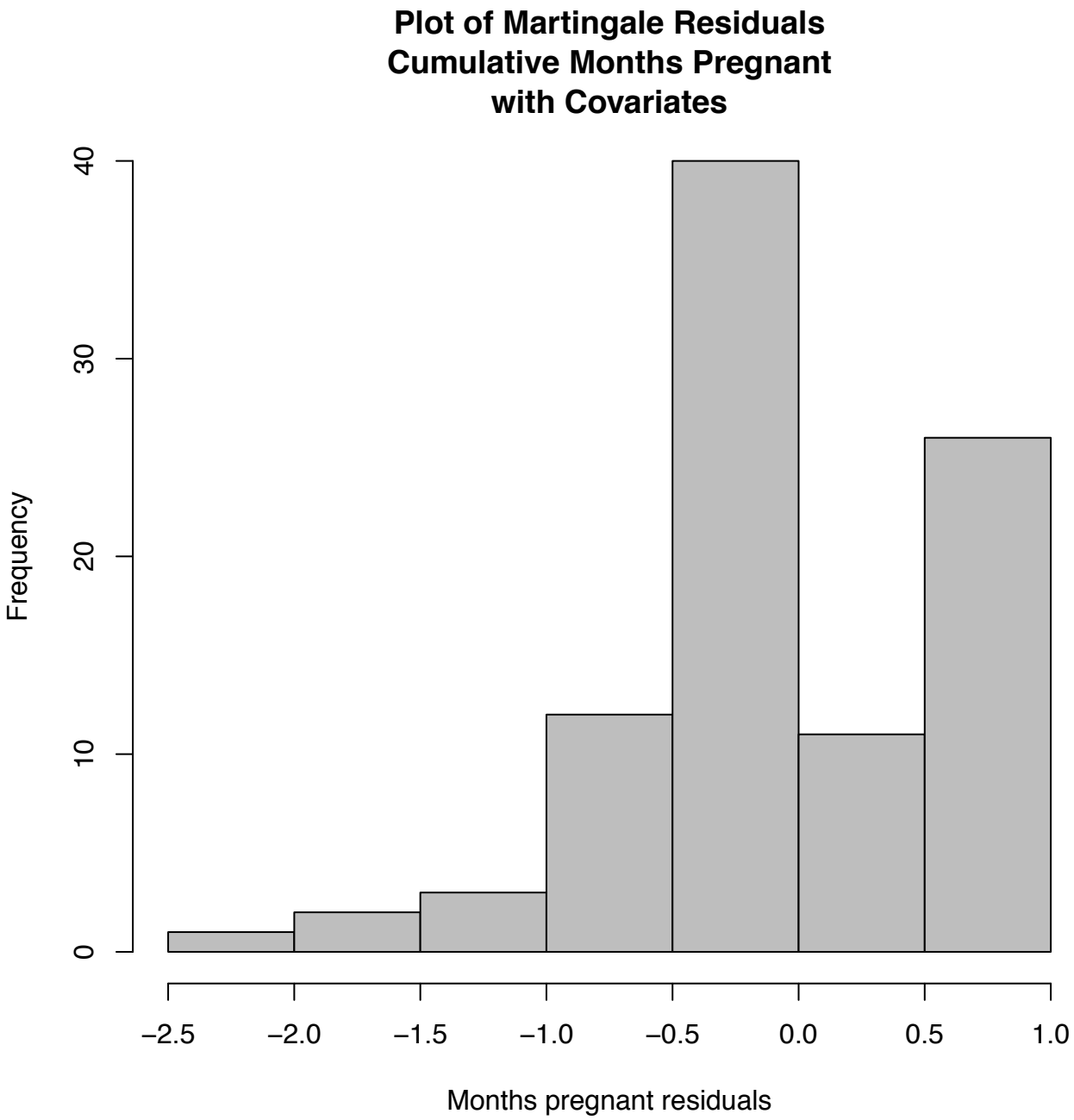

Figure S1. Martingale residuals for cumulative months pregnant Cox regression.

Cox model testing whether months pregnant in lifetime pregnant influences risk of AD onset. Consideration of the martingale residuals is useful for discerning the legitimacy of the Cox models fitted. When Martingale residuals histograms reveal a tight distribution and no outlier cases, this indicates that the model is a good reflection of trends in the data and not excessively influenced by a small number of individual cases. The distribution here reveals that no particular case or set of cases has undue influence on the model.

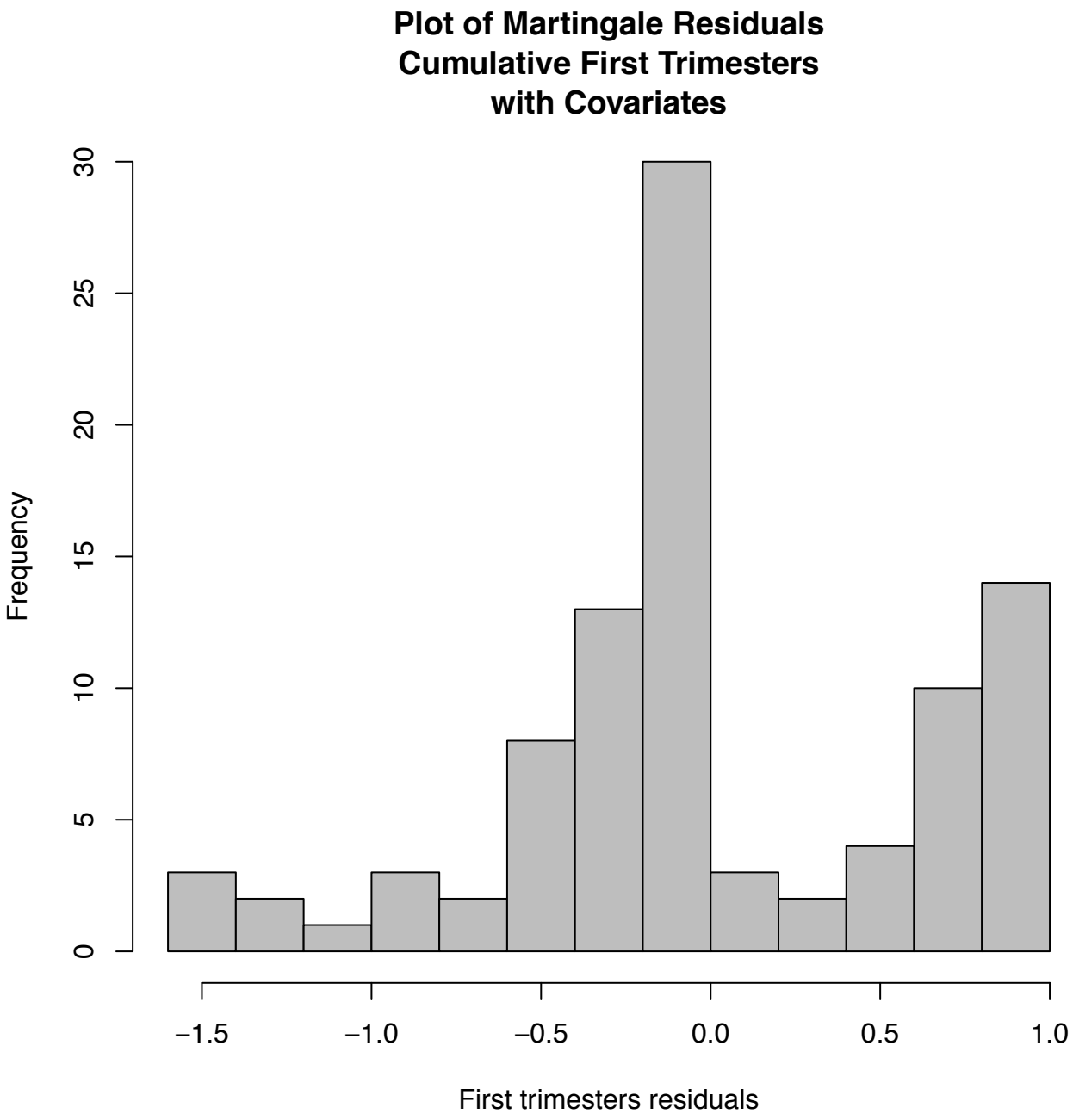

Figure S2. Martingale residuals for cumulative number of first trimesters Cox regressions

Cox model testing whether cumulative number of first trimesters influences risk of AD onset. Consideration of the martingale residuals is useful for discerning the legitimacy of the Cox models fitted. When Martingale residuals histograms reveal a tight distribution and no outlier cases, this indicates that the model is a good reflection of trends in the data and not excessively influenced by a small number of individual cases. The distribution here reveals that no particular case or set of cases has undue influence on the model.