endometriosis and control groups were strictly defined according to the r-AFS classification. By this means, we could apply the best definition of cases and controls. In the previous study, control groups consisted primarily of asymptomatic volunteers. Without surgical diagnosis, the main concern is that the control population could contain a substantial number of women with undiagnosed endometriosis, thereby diluting the risk factor effects (29). Secondly, these studies differed in the study participants. It is known that pregnancy and lactation can cause disease regression and improvements in symptoms of endometriosis (30-31). To reflect the authentic phenotype of endometriosis at the time of surgical diagnosis, our study excluded women who had ever given birth or lactated. Lastly, considerable intraobserver and interobserver variability is reported in r-AFS classification (32). Although this variability is a concern in any case-control study of endometriosis, it does not significantly decrease the validity or reliability of the association found.

This study provides evidence for an association between the *AhRR* codon 185 polymorphism and endometriosis, although the exact mechanisms for this effect are still unknown. One possible explanation of this association is alteration of AhR-mediated signaling by the polymorphism. The *AhRR* C/G + G/G genotype may facilitate proliferation of endometrial cells through the diminished down-regulation of

AhR-mediated signaling. Because of the small sample size, this is not a conclusive study, and these results need to be investigated further in large studies that also consider ethnic variation.

In summary, *AhRR* codon 185 polymorphism is associated with susceptibility to and severity of endometriosis in Japanese women. *AhR* codon 554 and *ARNT* codon 189 polymorphisms appeared not to be associated with endometriosis. The pathogenesis of endometriosis is still not clearly understood. The *AhRR* codon 185 polymorphism could be a useful genetic marker to predict the susceptibility to and severity of endometriosis.

ACKNOWLEDGEMENTS

We thank Tomoyuki Hanaoka, MD, PhD, Epidemiology and Prevention Division, Research Center for Cancer Prevention and Screening, National Cancer Center, for helpful discussions and Hiroyuki Nakao, PhD, Department of Public Health, Miyazaki Medical College, University of Miyazaki, for statistical advice and analysis.

This work was supported in part by a Grant-in-Aid for Risk Analysis Research on Food and Pharmaceuticals and Cancer Research from the Ministry of Health, Labour and Welfare of Japan.

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Table 1 Primers and probes used for real-time PCR analysis

Primers	Sequence
AhR codon 554	AGA to AAA, Arg to Lys
Forward primer	5' -AAA AAC AGT GAC TTG TAC AGC ATA ATG A-3'
Reverse primer	5' -CTG AAG TCA ACC TCA CCA GAA AAA T-3'
Probe: G allele	5' -FAM-TGA AGA CAT CAG ACA CAT-MGB-3'
Probe: A allele	5' -VIC-AGA CAT CAA ACA CAT GC-MGB-3'
ARNT codon 189	GTG to GTC, silent mutation
Forward primer	5' -TGC TGC CAA ACC ATT CAG ACT-3'
Reverse primer	5' -GGA ACT GAA ACA TTT GAT CTT GGA-3'
Probe: G allele	5' -VIC-CGG AGT CAG ACA CAT A-MGB-3'
Probe: C allele	5' -FAM-ACG GAG TCA GAG ACA T-MGB-3'
AhRR codon 185	CCC to GCC, Pro to Ala
Forward primer	5' -AGA CGG ATG TAA TGC ACC AGA A-3'
Reverse primer	5' -AGA GGC AGC GAT GTG TTA TGG-3'
Probe: C allele	5' -FAM-TGG GCA GCC CCC CGC C-TAMRA-3'
Probe: G allele	5' -VIC-TGG GCA GGC CCC GCC -TAMRA-3'

Table 2 Genotype and allele frequencies of AhR, ARNT, AhRR polymorphisms

Polymorphisms	Codons	Amino acids	Genotype frequencies		Allele frequencies	
			Endometriosis n (%)	Controls n (%)	Endometriosis n (%)	Controls n (%)
AhR codon 554						
G/G	AGA/AGA	Arg/Arg	24 (30.4)	22 (37.3)	G: 83 (52.5)	G: 73 (61.9)
A/G	AAA/AGA	Arg/Lys	35 (44.3)	29 (49.1)		
A/A	AAA/AAA	Lys/Lys	20 (25.3)	8 (13.6)	A: 75 (47.5)	A: 45 (38.1)
ARNT codon 189						
G/G	GTG/GTG	Val/Val	26 (32.9)	19 (32.2)	G: 92 (58.2)	G: 64 (54.2)
C/G	GTC/GTG	Val/Val	40 (50.6)	26 (44.1)		
C/C	GTC/GTC	Val/Val	13 (16.5)	14 (23.7)	C: 66 (41.8)	C: 54 (45.8)
AhRR codon 185						
C/C	CCC/CCC	Pro/Pro	20 (25.3)	27 (45.8)	C: 87 (55.1)	C: 81 (68.6)
C/G	CCC/GCC	Pro/Ala	47 (59.5)	27 (45.8)		
G/G	GCC/GCC	Ala/Ala	12 (15.2)	5 (8.4)	G: 71 (44.9)	G: 37 (31.4)

Table 3 AhR, ARNT and AhRR polymorphisms and risk of endometriosis

Polymorphism	Endometriosis n (%)	Controls n (%)	Crude OR (95% CI)	Adjusted OR[a] (95% CI)
AhR codon 554				
G/G	24 (30.4)	22 (37.3)	1	1
A/G + A/A	55 (69.6)	37 (62.7)	1.36 (0.67-2.78)	1.65 (0.76-3.61)
ARNT codon 189				
G/G	26 (32.9)	19 (32.2)	1	1
C/G + C/C	53 (67.1)	40 (67.8)	0.97 (0.47-1.99)	0.86 (0.39-1.87)
AhRR codon 185				
C/C	20 (25.3)	27 (45.8)	1	1
C/G + G/G	59 (74.7)	32 (54.2)	2.49* (1.21-5.12)	2.53* (1.16-5.55)

[a]ORs adjusted for age and menstrual characteristics.

*P < 0.05

Table 4 AhRR codon 185 polymorphism and severity of endometriosis

Clinical stage	AhRR C/C genotype	AhRR C/G + G/G genotype	Crude OR (95% CI)	Adjusted OR[a] (95% CI)
Controls n (%)	27 (45.8)	32 (54.2)	1	1
Stage I - II n (%)	8 (25.8)	23 (74.2)	2.43 (0.94-6.30)	1.78 (0.64-4.98)
Stage III-IV n (%)	12 (25.0)	36 (75.0)	2.53* (1.10-5.81)	3.17* (1.27-7.91)

[a]ORs adjusted for age and menstrual characteristics.

*P < 0.05

P for trend: 0.02