Journal of Clinical Laboratory Analysis 24 : 182–186 (2010)

Evaluation of the Poly(ADP-ribose) Polymerase-1 Gene Variants in Alzheimer's Disease

Hsin-Ping Liu,[1] Wei-Yong Lin,[2,3] Bor-Tsang Wu,[4] Shu-Hsiang Liu,[5] Wen-Fu Wang,[6] Chon-Haw Tsai,[7,8] Chun-Cheng Lee,[3,4] and Fuu-Jen Tsai[3,5,9]*

[1]*Graduate Institute of Acupuncture Science, China Medical University, Taichung, Taiwan*
[2]*Graduate Institute of Integrated Medicine, China Medical University, Taichung, Taiwan*
[3]*Department of Medical Research, China Medical University Hospital, Taichung, Taiwan*
[4]*School of Physical Therapy, China Medical University, Taichung, Taiwan*
[5]*Department of Biotechnology, Asia University, Taichung, Taiwan*
[6]*Department of Neurology, Chang-Hua Christian Hospital, Chang-Hua, Taiwan*
[7]*Graduate Institute of Neural and Cognitive Sciences, China Medical University, Taichung, Taiwan*
[8]*Departments of Neurology, China Medical University Hospital, Taichung, Taiwan*
[9]*Graduate Institute of Chinese Medical Science, China Medical University, Taichung, Taiwan*

Amyloid peptide is thought to play a critical role in neuronal death in Alzheimer's disease (AD), most likely through oxidative stress. Free radical-related injury leads to DNA breaks, which subsequently activates the repair enzyme poly(ADP-ribose) polymerase-1 (PARP-1). In this study, the relationship between genetic variants situated at the *PARP-1* gene and AD development was investigated. We performed a case and control study from a Taiwanese population enrolled 120 AD patients and 111 healthy controls by using a polymerase chain reaction restriction fragment length polymorphism approach for two *PARP-1* exonic polymorphisms, 414C/T (rs1805404) and 2456T/C (rs1136410), corresponding to protein residues at positions 81Asp/Asp and 762Val/Ala. There were no significant differences in allele or genotype frequencies for either *PARP-1* gene variant between the case and control groups; however, upon analysis of the haplotype distribution, four haplotypes (Hts) were identified. We found that the distributions of Ht3-TT and Ht4-CC were significantly associated with an increased risk of AD ($P<0.0001$), whereas the Ht1-TC haplotype showed a protective effect for cases compared with the control group ($P<0.05$). These results reveal that the *PARP-1* gene is highly associated with AD susceptibility and might contribute to a critical mechanism that mediates cell survival or death as a response to cytotoxic stress. J. Clin. Lab. Anal. 24:182–186, 2010.

Key words: Alzheimer's disease; PARP-1; polymorphism

INTRODUCTION

Alzheimer's disease (AD) has long been known to be an age-related disorder with several clinical symptoms, including neurodegenerative processes, cognitive dysfunction, and dementia. Two neuropathological hallmarks that appear in the brain region of AD patients are major aggregates of senile plaques and neurofibrillary tangles, which are composed of amyloid peptides (Aβ) and hyperphosphorylated Tau proteins, respectively (1,2). Mutations in genes coding for amyloidgenic processing, including amyloid precursor protein, presenilin 1 and 2, have been found to predominantly lead to the production of amyloid plaques (3–5); however, as

Grant sponsor: Taiwan Department of Health Clinical Trial and Research Center of Excellence; Grant number: D0H99-TD-B111-004; Grant sponsor: China Medical University and Hospital; Grant numbers: CMU97-CMC-010; CMU 96-264, DMR-97-125.

Hsin-Ping Liu and Wei-Yong Lin contributed equally to this work.

*Correspondence to: Fuu-Jen Tsai, Department of Medical Research, China Medical University Hospital, No. 2 Yuh Der Road, Taichung 404, Taiwan. E-mail: d0704@www.cmuh.org.tw

Received 7 January 2010; Accepted 19 March 2010
DOI 10.1002/jcla.20379
Published online in Wiley InterScience (www.interscience.wiley.com).

most cases are sporadic and have unclear causes, this suggests that additional genetic factors responsible for AD progression still need to be identified.

The accumulation of reactive oxygen species (ROS), which leads to marked increases in oxidative stress, is thought to be a probable mechanism in the pathogenesis of several diseases, including ischemia, human prion disease as well as in AD patients (6–8). Excessive Aβ accumulation is also correlated with the free radical production and impairment of neuronal survival in transgenic mouse models of AD (9,10). This implies that overproduction of Aβ and their assembly into aggregated forms may appear to cause potent neurotoxicity and lead to the disturbance of neurotransmission and even advanced cognitive behavior, contributing to the unique AD etiology (11,12).

In brain sections of AD patients, poly(ADP-ribose) polymerase-1 (PARP-1) overactivation and accumulation of its end-modification, poly(ADP-ribosyl)ation were observed by immunohistochemical analysis when compared with age-matched controls (13). PARP-1 is a nuclear enzyme and plays an important role in mediating the poly(ADP-ribose) reaction at target proteins after cells encounter excitotoxicity and free radical-induced damage in an energy depletion process through cytosolic NAD^+ (14,15). Aβ deposits or other inducers could initiate oxidative stress and DNA strand breaks that serve as signals to stimulate PARP-1 activity, thus upregulated levels of functional PARP-1 are strongly related to cell death processes that are relevant to neurodegenerative disease (16). To explore the possible effects of genetic risk for the preapoptotic protein PARP-1 with respect to susceptibility to AD pathophysiological processes, we performed a case and control study. Our results show an associative relationship for the *PARP-1* gene, which may have profound implications as a genetic risk marker in a Taiwanese population with AD progression.

MATERIALS AND METHODS

Subjects

A total of 120 cases with late-onset AD (mean age at onset, 74.9 years; SD 6.7, 54.2% female) and 111 age-related normal control group subjects (mean age 67.1 years; SD 8.3, 64% female) were recruited for this study. Participants were enrolled from Chang-Hua Christian Hospital and China Medical University Hospital. This project was approved by an institutional ethics committee and informed consent was obtained from all subjects. The diagnosis of AD was determined clinically according to the guidelines of the National Institute of Neurological and Communicative Disorders and Stroke-Alzheimer's Disease and Related Disorders Association (NINCDS–ADRDA). Dementia was diagnosed on the basis of Diagnostic and Statistical Manual of Mental Disorders criteria.

Determination of *PARP-1* Gene Variants

For the analysis of *PARP-1* gene variants, two single nucleotide polymorphisms (SNPs) across the exonic *PARP-1* locus were selected for this study from the public dbSNP database: 414C/T (rs1805404) and 2456T/C (rs1136410), encoding protein residues at positions 81Asp/Asp and 762Val/Ala, respectively. In order to identify allele preference, genomic DNA that was extracted from peripheral blood samples through standard protocols (Genomic DNA kit, Qiagen, Valencia, CA) was chosen for genotyping. Experiments were performed on the basis of a PCR-restriction fragment length polymorphism (PCR-RFLP) methodology. Specific primers and restriction enzymes were used for each reaction (Table 1). Briefly, PCR reactions were carried out in a total volume of 25 μl containing 5 ng genomic DNA and forward and reverse primers. The PCR amplification protocol was set as 95°C for 5 min, followed by 40 cycles of 95°C for 30 sec, the specific Tm of each paired-primer for 30 sec, 72°C for 40 sec, and a final elongation step at 72°C for 7 min. Five microliters of PCR amplicons were analyzed by different restriction enzyme digests (New England Biolabs, Ont., Canada) at 37°C overnight in a total volume of 20 μl. PCR products and digestion fragments were determined by agarose gel electrophoresis and were visualized directly with ethidium bromide.

Statistical Analysis

The distributions of allele and genotype frequencies were performed for each genotyping dataset by the χ^2

TABLE 1. Primers of *PARP-1* Gene Polymorphisms Performed by PCR-RFLP

Polymorphisms	Primers	Restriction enzyme	Alleles	Allele size (bp)	refSNP no.
414 C/T	F: 5′-AAGGTCTAGTGGGTCTAAGTCA-3'	DpnII	C	319	rs1805404
	R: 5′-AGGTTTGCTTTGCTCTCTGAGA-3'		T	(187+132)	
2456 T/C	F: 5′-GTACGAGAGGAAAGACAGTTCT-3'	AciI	T	388	rs1136410
	R: 5′-CCTGACCCTGTTACCTTAATGT-3′		C	(253+135)	

test. Statistical analyses of the odds ratios (OR) and 95% confidence interval (CI) were carried out with SPSS version 10.0 software (Chicago, IL), based on the presence of the reference allele and genotype frequencies. Results were considered significant only when the *P* value was less than 0.05. Adherence to the Hardy–Weinberg equilibrium constant was tested using the χ^2 test with one degree of freedom. The haplotype approach was analyzed from unphased genotype data in the program Phase v2.1 algorithm, according the Bayesian statistical method (17,18).

RESULTS

The *PARP-1* gene 414C/T synonymous variant (rs1805404) at codon 81Asp/Asp and the 2456T/C polymorphism (rs1136410) causing the codon change 762Val/Ala were assayed by PCR-RFLP. Genomic DNA samples from AD cases and healthy controls were prepared for PCR reactions. Table 1 shows the specific primers and restriction enzymes for each PCR preparation and genotyping analysis. While performing PCR-RFLP, products were identified as digestible or indigestible homozygous "TT" and "CC" genotypes as well as the heterozygous "CT" genotype for the *PARP-1* gene polymorphism 414C/T using the restriction enzyme *DpnII*. Genotyping of the 2456T/C variant was performed by *AciI* treatment; the "T" allele remained intact compared with the two DNA fragments found in the "C" allele sequence.

Table 2 shows the distributions of allele and genotype frequencies for *PARP-1* gene polymorphisms in AD cases and controls. For the 414C/T gene variant, the allele frequency of C:T in patients is similar to that of the control group (52.9:47.1% in cases vs. 59.5:40.5% in the control group), as was the genotype frequency; thus, no significant difference was observed between AD cases and healthy controls ($P > 0.05$). With the 2456T/C *PARP-1* gene polymorphism, the distribution of the allele frequency for T:C was 52.2:47.8% in the AD group vs. 59.2:40.8% in the control group. Neither the T/C allele distribution nor the TT/TC/CC genotype frequency was significantly associated with either the case or the control group ($P > 0.05$).

We analyzed the association of haplotype distributions between patient cases and the control group. Haplotype frequencies of the *PARP-1* gene at two polymorphisms appear in Table 3, with four haplotypes observed in our survey. The frequency of the most common haplotype (Ht1-TC) was 42.9% in patients compared with 59.0% in controls. Two haplotypes, Ht3-TT and Ht4-CC, appeared much more frequently in patients than in the

TABLE 2. Genotype and Allele Frequencies of *PARP-1* Gene Polymorphisms Between Patients and Healthy Controls

Polymorphisms		Cases number (%)	Controls number (%)	*P*-value[a]/Cp-value[b]	Odds ratio (95% CI)
414 C/T	CC	33 (27.7)	38 (34.5)	0.3495/0.6990	0.57 (0.26–1.23)
	CT	60 (50.4)	55 (50.0)		0.71 (0.35–1.45)
	TT	26 (21.8)	17 (15.5)		1
	C	126 (52.9)	131 (59.5)	0.1547/0.3094	0.76 (0.53–1.11)
	T	112 (47.1)	89 (40.5)		1
2456 T/C	TT	33 (28.4)	37 (33.9)	0.2589/0.5178	0.54 (0.25–1.16)
	TC	55 (47.4)	55 (50.5)		0.61 (0.30–1.23)
	CC	28 (24.1)	17 (15.6)		1
	T	121 (52.2)	129 (59.2)	0.1343/0.2686	0.75 (0.52-1.09)
	C	111 (47.8)	89 (40.8)		1

CI, confidence interval. Hardy–Weinberg equilibrium: 414C/T ($P = 0.897$ and 0.692 for cases and controls, respectively); 2456T/C ($P = 0.591$ and 0.643 for cases and controls, respectively).
[a]*P*-value was compared by χ^2 test.
[b]Cp-value: *P*-values corrected by Bonferroni correction.

TABLE 3. Haplotypes of *PARP-1* Gene Polymorphisms for the Association Between AD Patients and Controls

Haplotypes	414C/T	2456T/C	Cases (%)	Controls (%)	*P*-value[a]/Cp-value[b]	Odds ratio (95% CI)
Ht1	T	C	103 (42.9)	131 (59.0)	0.0005/0.0020	0.52 (0.36–0.76)
Ht2	C	T	89 (37.1)	89 (40.0)	0.5070/2.0280	0.88 (0.61–1.28)
Ht3	T	T	24 (10.0)	2 (1.0)	$8.7174 \times 10^{-6}/3.487 \times 10^{-5}$	12.22 (2.85–52.35)
Ht4	C	C	24 (10.0)	0 (0.0)	$9.4989 \times 10^{-8}/3.800 \times 10^{-7}$	

[a]*P*-value was compared by χ^2 test
[b]Cp-value: *P*-values corrected by Bonferroni correction., CI, confidence interval.

control group. After a haplotype-specific analysis, the haplotype Ht1-TC appeared to be a significantly more "protective" haplotype (Ht1-TC, $P = 0.002$, OR: 0.52, 95% CI: 0.36–0.76) than the non-Ht1 haplotype among AD patients and the control group. A similar haplotype-specific analysis showed that Ht3-TT and Ht4-CC appeared to be significant "risk" haplotypes (Ht3-TT, $P = 3.5 \times 10^{-5}$, OR: 12.22, 95% CI: 2.85–52.35; Ht4-CC, $P = 3.8 \times 10^{-7}$) between the two groups. Thus, subjects with the "Ht1-TC" haplotype of the *PARP-1* gene 414C/T and 2456T/C polymorphisms normally exhibited lower incidences of pathogenic AD; in contrast, subjects with the "Ht3-TT" or "Ht4-CC" haplotype appear to be at higher risk to develop the disease.

DISCUSSION

Clinically, different brain regions, such as the hippocampus and cerebral cortex, have shown irreversible neuronal loss in AD patients and this neurodegenerative process causes severe brain dysfunction in an age-dependent manner. We suggested that oxidative stress and DNA damage might be a relevant event for this pathology. Thus, we tested our proposal to determine whether the *PARP-1* gene is associated with AD incidence.

Poly(ADP-ribosyl)ation is an important modulator in the response to cellular injuries such as DNA breaks, excitotoxicity, and oxidative stress (14,19). Changes in poly(ADP-ribosyl)ation may trigger downstream signaling that either facilitate DNA repair or commit to a cell death program (20) and increased levels of poly(ADP-ribosyl)ation present a crucial pathogenic phenomenon in the patients with conditions such as stroke, cancer, and neurodegenerative diseases (21–23). Oxidative stress and neuronal damage in AD patients may be primarily caused by the assembly states of Aβ and subsequent activation of NMDA receptors (24,25). NMDA receptors mediate excitotoxicity through excess Ca^{2+} influx, and the formation of nitric oxide and highly ROS lead to PARP-1 activation (22,26); this process can be blocked by a decrease in PARP-1 protein expression or by PARP-1 inhibitors (27). Similar result has also been observed in the adult rat hippocampus, indicating that mediation of PARP-1 activity is a critical cellular mechanism that regulates cell viability or death in response to stress and cytotoxicity insults (28).

According to several clinical studies, accumulated levels of PARP-1 protein and poly(ADP-ribose) reaction have been detected not only in brain tissue, but also in peripheral blood mononuclear cells from AD patients compared with healthy subjects (13,29,30), suggesting that activation of PARP-1 presents a positive association between Aβ overproduction and oxidative stress-related damage in AD progression. Therefore, to explore the association between *PARP-1* gene variants and the incidence of disease development, two polymorphic variants located in the *PARP-1* coding region were examined. One is a synonymous SNP at C/T, situated at position 414 (Asp81Asp), whereas the other is a nonsynonymous T/C substitution, located at position 2456, which causes a valine-to-alanine change in codon 762 (Val762Ala). In our analysis of these two *PARP-1* gene variants, no significant differences were observed between AD cases and healthy controls in allele or genotype frequency ($P > 0.05$). However, after an associative analysis of haplotype distributions, patients with haplotypes Ht3-TT and Ht4-CC presented with an increased risk of disease development (both $P < 0.0001$); on the contrary, Ht1-TC appeared to be a significantly "protective" haplotype between cases and the control group ($P = 0.002$). As we evaluated according to the Better Associations for Disease and GEnes system for describing genetic associations, this is a second-class association ($P < 1 \times 10^{-4}$), and is therefore expected the association belonged to higher relativity (31). Because this is a Class 2 association and is therefore expected to be reproducible, our finding can be expected to reproduce in spite of the lack of replication.

Several reports have demonstrated that Val762Ala lies within the catalytic domain of the PARP-1 protein, and replacing valine with alanine causes a decrease in PARP-1 enzyme activity (32,33). This variant was found to be associated with several diseases, including prostate cancer, thyroid carcinoma, and systemic lupus erythematosus (32,34,35). In addition, *PARP-1* gene variants at the promoter region also had significant correlations with the development of Parkinson's disease as well as AD in a Spanish population (36,37). PARP-1 activation may be an important sensor for the initiation of cellular death processes and may mediate the balance of cellular survival and death processes. This study investigates the polymorphic association of the *PARP-1* gene to patients with AD in a Taiwanese population. Our results may provide evidence for a genetic risk factor that could be used to enhance the prediction of AD progression.

REFERENCES

1. Masters CL, Simms G, Weinman NA, Multhaup G, McDonald BL, Beyreuther K. Amyloid plaque core protein in Alzheimer disease and Down syndrome. Proc Natl Acad Sci U S A 1985;82:4245–4249.
2. Goedert M, Wischik CM, Crowther RA, Walker JE, Klug A. Cloning and sequencing of the cDNA encoding a core protein of the paired helical filament of Alzheimer disease: identification as the microtubule-associated protein tau. Proc Natl Acad Sci U S A 1988;85:4051–4055.
3. Mullan M, Crawford F, Axelman K, et al. A pathogenic mutation for probable Alzheimer's disease in the APP gene at the N-terminus of beta-amyloid. Nat Genet 1992;1:345–347.

4. Sherrington R, Rogaev EI, Liang Y, et al. Cloning of a gene bearing missense mutations in early-onset familial Alzheimer's disease. Nature 1995;375:754–760.
5. Steiner H, Romig H, Grim MG, et al. The biological and pathological function of the presenilin-1 Deltaexon 9 mutation is independent of its defect to undergo proteolytic processing. J Biol Chem 1999;274:7615–7618.
6. Niizuma K, Endo H, Chan PH. Oxidative stress and mitochondrial dysfunction as determinants of ischemic neuronal death and survival. J Neurochem 2009;109:133–138.
7. Kovacs GG, Kalev O, Budka H. Contribution of neuropathology to the understanding of human prion disease. Folia Neuropathol 2004;42:69–76.
8. Reddy PH. Amyloid precursor protein-mediated free radicals and oxidative damage: implications for the development and progression of Alzheimer's disease. J Neurochem 2006;96:1–13.
9. Smith MA, Hirai K, Hsiao K, Pappolla MA, et al. Amyloid-beta deposition in Alzheimer transgenic mice is associated with oxidative stress. J Neurochem 1998;70:2212–2215.
10. Schuessel K, Schafer S, Bayer TA, et al. Impaired Cu/Zn-SOD activity contributes to increased oxidative damage in APP transgenic mice. Neurobiol Dis 2005;18:89–99.
11. Kim JH, Anwyl R, Suh YH, Djamgoz MB, Rowan MJ. Use-dependent effects of amyloidogenic fragments of (beta)-amyloid precursor protein on synaptic plasticity in rat hippocampus in vivo. J Neurosci 2001;21:1327–1333.
12. LaFerla FM, Green KN, Oddo S. Intracellular amyloid-beta in Alzheimer's disease. Nat Rev Neurosci 2007;8:499–509.
13. Love S, Barber R, Wilcock GK. Increased poly(ADP-ribosyl)ation of nuclear proteins in Alzheimer's disease. Brain 1999;122:247–253.
14. Ame JC, Spenlehauer C, de Murcia G. The PARP superfamily. Bioessays 2004;26:882–893.
15. Lindahl T, Satoh MS, Poirier GG, Klungland A. Post-translational modification of poly(ADP-ribose) polymerase induced by DNA strand breaks. Trends Biochem Sci 1995;20:405–411.
16. Szabo C, Zingarelli B, O'Connor M, Salzman AL. DNA strand breakage, activation of poly (ADP-ribose) synthetase, and cellular energy depletion are involved in the cytotoxicity of macrophages and smooth muscle cells exposed to peroxynitrite. Proc Natl Acad Sci U S A 1996;93:1753–1758.
17. Stephens M, Donnelly P. A comparison of bayesian methods for haplotype reconstruction from population genotype data. Am J Hum Genet 2003;73:1162–1169.
18. Stephens M, Smith NJ, Donnelly P. A new statistical method for haplotype reconstruction from population data. Am J Hum Genet 2001;68:978–989.
19. Weinfeld M, Chaudhry MA, D'Amours D, et al. Interaction of DNA-dependent protein kinase and poly(ADP-ribose) polymerase with radiation-induced DNA strand breaks. Radiat Res 1997;148:22–28.
20. Tulin A, Spradling A. Chromatin loosening by poly(ADP)-ribose polymerase (PARP) at Drosophila puff loci. Science 2003;299: 560–562.
21. Pacher P, Beckman JS, Liaudet L. Nitric oxide and peroxynitrite in health and disease. Physiol Rev 2007;87:315–424.
22. Koh DW, Dawson TM, Dawson VL. Poly(ADP-ribosyl)ation regulation of life and death in the nervous system. Cell Mol Life Sci 2005;62:760–768.
23. Canan Koch SS, Thoresen LH, Tikhe JG, et al. Novel tricyclic poly(ADP-ribose) polymerase-1 inhibitors with potent anticancer chemopotentiating activity: design, synthesis, and X-ray cocrystal structure. J Med Chem 2002;45:4961–4974.
24. Mucke L, Masliah E, Yu GQ, et al. High-level neuronal expression of abeta 1-42 in wild-type human amyloid protein precursor transgenic mice: Synaptotoxicity without plaque formation. J Neurosci 2000;20:4050–4058.
25. Tanzi RE, Bertram L. Twenty years of the Alzheimer's disease amyloid hypothesis: a genetic perspective. Cell 2005; 120:545–555.
26. Zhang J, Dawson VL, Dawson TM, Snyder SH. Nitric oxide activation of poly(ADP-ribose) synthetase in neurotoxicity. Science 1994;263:687–689.
27. Yu SW, Wang H, Poitras MF, et al. Mediation of poly(ADP-ribose) polymerase-1-dependent cell death by apoptosis-inducing factor. Science 2002;297:259–263.
28. Strosznajder JB, Jesko H, Strosznajder RP. Effect of amyloid beta peptide on poly(ADP-ribose) polymerase activity in adult and aged rat hippocampus. Acta Biochim Pol 2000;47:847–854.
29. Kassner SS, Bonaterra GA, Kaiser E, et al. Novel systemic markers for patients with Alzheimer disease?—a pilot study. Curr Alzheimer Res 2008;5:358–366.
30. Dorszewska J, Florczak J, Rozycka A, Jaroszewska-Kolecka J, Trzeciak WH, Kozubski W. Polymorphisms of the CHRNA4 gene encoding the alpha4 subunit of nicotinic acetylcholine receptor as related to the oxidative DNA damage and the level of apoptotic proteins in lymphocytes of the patients with Alzheimer's disease. DNA Cell Biol 2005;24:786–794.
31. Manly KF. Reliability of statistical associations between genes and disease. Immunogenetics 2005;57:549–558.
32. Lockett KL, Hall MC, Xu J, et al. The ADPRT V762A genetic variant contributes to prostate cancer susceptibility and deficient enzyme function. Cancer Res 2004;64:6344–6348.
33. Wang XG, Wang ZQ, Tong WM, Shen Y. PARP1 Val762Ala polymorphism reduces enzymatic activity. Biochem Biophys Res Commun 2007;354:122–126.
34. Chiang FY, Wu CW, Hsiao PJ, et al. Association between polymorphisms in DNA base excision repair genes XRCC1, APE1, and ADPRT and differentiated thyroid carcinoma. Clin Cancer Res 2008;14:5919–5924.
35. Hur JW, Sung YK, Shin HD, Park BL, Cheong HS, Bae SC. Poly(ADP-ribose) polymerase (PARP) polymorphisms associated with nephritis and arthritis in systemic lupus erythematosus. Rheumatology (Oxford) 2006;45:711–717.
36. Infante J, Sanchez-Juan P, Mateo I, et al. Poly (ADP-ribose) polymerase-1 (PARP-1) genetic variants are protective against Parkinson's disease. J Neurol Sci 2007;256:68–70.
37. Infante J, Llorca J, Mateo I, et al. Interaction between poly(ADP-ribose) polymerase 1 and interleukin 1A genes is associated with Alzheimer's disease risk. Dement Geriatr Cogn Disord 2007;23: 215–218.