

Epidemiology / Épidémiologie

# Interaction between genes and environment in neurodegenerative diseases

Alexis Elbaz, Carole Dufouil, Annick Aléproux \*

*Inserm, U708 'Neuroépidémiologie' & université Pierre-et-Marie-Curie (Paris-6), groupe hospitalier Pitié-Salpêtrière,  
75651 Paris cedex 13, France*

Received 11 January 2007; accepted after revision 14 February 2007

Available online 9 April 2007

Presented by Alain-Jacques Valleron

## Abstract

Alzheimer's disease (AD) and Parkinson's disease (PD) are highly prevalent disorders that account for a large part of the global burden of neurodegenerative diseases. Most AD and PD cases occur sporadically and it is generally agreed that they could arise through interactions among genetic and environmental factors. Candidate genes involved in the metabolism of xenobiotics, neurodegeneration and functioning of dopaminergic neurons were found to be associated with PD. Some of these genes interact with environmental factors that could modify PD risk. Thus, we found that the inverse association between smoking and the risk of PD depended on a polymorphism of the iNOS (inducible NO synthase) gene. We also found that the cytochrome P450 2D6 gene could have a modifying effect on the risk of PD among persons exposed to pesticides. Both interactions have biological plausibility supported by laboratory studies and could contribute to better understand the aetiology of PD. A single susceptibility gene has been identified in sporadic AD. The  $\epsilon 4$  allele of epsilon polymorphism of the apolipoprotein E gene (APOE) is strongly associated with AD, the risk of AD being multiplied by 5 in persons carrying two  $\epsilon 4$  alleles. The mechanism of the association between APOE and AD is poorly understood. A few interactions between the epsilon polymorphism and possible risk factors for AD have been described. However, these interactions had no biological plausibility and were likely due to chance. **To cite this article:** A. Elbaz *et al.*, *C. R. Biologies 330 (2007)*.

© 2007 Académie des sciences. Published by Elsevier Masson SAS. All rights reserved.

**Keywords:** Alzheimer disease; Parkinson disease; Epidemiology; Gene; Environment

## 1. Introduction

According to the Global Burden of Disease Study (GBD), a collaborative study of the World Health Organization (WHO), the World Bank and the Harvard School of Public Health, dementia and other neurodegenerative diseases will be, in 2020, the eighth cause of

disease burden for developed regions [1,2]. The GBD researchers have also weighted the severity of disability for a series of health conditions. Out of the ten disorders within the three highest disability classes, eight are neurological problems. According to the WHO, neurodegenerative diseases will become the world's second leading cause of death by the middle of the century, overtaking cancer [2]. Although such rough rankings and predictions are questionable, they confirm that neurodegenerative diseases are an increasing public con-

\* Corresponding author.

E-mail address: [alperovi@chups.jussieu.fr](mailto:alperovi@chups.jussieu.fr) (A. Aléproux).

cern. AD, other ageing-associated dementias, and PD account currently for a large part of the global burden of neurodegenerative diseases. This part is destined to increase markedly worldwide because of demographic evolution and epidemiological transition, unless more effective preventive procedures or treatments are available. In France, the numbers of AD and PD cases are currently estimated at 800 000 and 100 000, respectively. The main epidemiological characteristics of these two diseases will be briefly described in the following sections of this paper.

Most neurodegenerative diseases are characterized by the aggregation of intracellular proteins: tau and amyloid in AD, synuclein in PD, prion protein in Creutzfeldt–Jakob disease, etc. [3]. Studies of familial forms of neurodegenerative diseases have shown that, in some families, the disease is due to a mutation of the gene coding for the abnormally aggregating protein. However, only a small minority of cases are of purely genetic origin. Most neurodegenerative disorders occur sporadically and they belong to the very long list of diseases that could arise through interactions among genetic and environmental factors. In the following paragraphs, we will describe a particular type of bias that may affect studies of genetic susceptibility in late-onset diseases. Then, we will present and discuss examples of gene–environment interactions in PD and AD.

## 2. Methodological difficulties in studies of genetic susceptibility and gene–environment interactions in neurodegenerative diseases

Studies of genetic susceptibility in neurodegenerative diseases (ND) are complicated by a number of issues due to their usually late age at onset. Exceptionally, ND can affect younger subjects; in such cases, the disease is often familial, and these families allow identifying single gene defects that provide important clues to pathophysiology. More commonly, the disease is sporadic and genetic susceptibility and gene–environment interactions are investigated using an association study design. The particularity of ND is that family-based designs cannot be easily used since parents and elder siblings are frequently not available for study. These difficulties explain that the most often-used study design has been the case-unrelated control design. Because it is likely that the size of the genetic effects involved is small, large studies are needed [4]. For detection of gene–environment interactions, even larger studies are needed and detailed assessment of environmental exposures can be difficult in that case.

Assessment of environmental exposures in studies of gene–environment interactions is a central question. It has been shown that error measurement in environmental exposures results in loss of power to detect interactions [5]. In addition, differential misclassification in exposure assessment can result in bias in the estimation of the interaction parameter, either toward or away from the null [6]. These issues are particularly relevant to ND. Neurodegeneration probably starts years before the clinical onset of ND, but the length of the pre-symptomatic period is poorly known; for some authors, it is even possible that very early exposures (e.g., in utero or during childhood) may affect the risk of subsequent ND. Thus, it is difficult to target a time period of interest for the assessment of environmental exposures, and they have to be assessed over very long time periods. In addition, because ND are often characterized by cognitive decline or dementia, recall of past exposures is even more difficult; proxies can be used, but they may not provide valid information for several types of exposures.

A particular type of bias may affect studies of genetic susceptibility in late-onset diseases [7–9]. ND usually occurs in elderly subjects who have survived to competing causes of death. These subjects may therefore have specific characteristics that should be taken into account when investigating risk factors for neurodegenerative diseases. For instance, the  $\epsilon 4$  allele of the epsilon polymorphism in the apolipoprotein E (apoE) gene is a risk factor for cardiovascular disease and its frequency decreases with age, likely due to cardiovascular deaths. Thus,  $\epsilon 4$  frequency is lower in elderly subjects at risk of neurodegenerative disease than in younger subjects, and age is an important confounder. In addition, if  $\epsilon 4$  interacts with an environmental risk factor to increase the risk of cardiovascular death, an association between  $\epsilon 4$  and the environmental risk factor will appear with increasing age and it should be taken into account when investigating the association between  $\epsilon 4$  and late-onset diseases such as ND.

More generally, let us consider a disease  $D_1$  (e.g., a cardiovascular disease) that results in high mortality and a disease  $D_2$  (e.g., ND) that occurs at a later age than  $D_1$ . Let  $E$  be an environmental risk factor and  $G$  the genetic susceptibility factor of interest (for simplicity we assume both variables to be dichotomous). Subscripts  $E$ ,  $G$  denote their presence and  $\bar{E}$ ,  $\bar{G}$  their absence. We assume that  $E$  and  $G$  are independent at time  $t_0$ , with initial frequencies  $P_{E(t_0)}$  and  $P_{G(t_0)}$ . The relative hazard (RH) of  $D_2$  associated with  $G$  at time  $t$  is

$$RH_{2G(t)} = \frac{RH_{2GE} P_{E/G(t)} + RH_{2G\bar{E}} P_{\bar{E}/G(t)}}{RH_{2\bar{G}E} P_{E/\bar{G}(t)} + P_{\bar{E}/\bar{G}(t)}}$$

where  $RH_{ij}$  is the relative hazard of  $D_i$  for the  $j^{\text{th}}$  category ( $i = 1, 2; j = \bar{G}\bar{E}, G\bar{E}, \bar{G}E, GE$ ) and  $P_{E/G(t)}$ ,  $P_{E/\bar{G}(t)}$ ,  $P_{\bar{E}/G(t)}$ , and  $P_{\bar{E}/\bar{G}(t)}$  are conditional frequencies of  $E$  given  $G$  at time  $t$ . It can be shown mathematically that  $RH_{2G(t)}$  varies with time and that  $E$  behaves as a confounder for the relation between  $G$  and  $D_2$  if (i)  $G$  and  $E$  interact on an additive scale for  $D_1$  and (ii)  $E$  is associated with  $D_2$ . Therefore, omitting  $E$  from the analysis when studying the relation between  $G$  and  $D_2$  results in a biased estimate of the relative hazard. The direction of the bias depends on the type of interaction (synergistic or antagonistic); over- or underestimation of the association can be observed and  $G$  can artificially appear as being associated with  $D_2$ . The importance of bias depends on the strength of the interaction between  $G$  and  $E$  for  $D_1$ , the cumulative incidence of  $D_1$ , and the initial frequency of  $G$  and  $E$ . Numerical examples show that, under certain conditions (strong interactions; high cumulative incidence of  $D_1$ ;  $E$  and  $G$  are neither rare nor very common), the bias can be important [9]. It may contribute to explain why studies carried out in populations with different characteristics may yield inconsistent results and why, in addition to aetiological heterogeneity, it is frequent to observe that the relative estimates associated with a genetic susceptibility factor decrease with age. For instance, the association between Alzheimer's disease and the  $\epsilon 4$  allele [10] and that between positive family history of PD and PD [11] strongly decrease with age.

### 3. Gene–environment interactions in Parkinson's disease

#### 3.1. Introduction

Parkinson's disease (PD) is the most common cause of Parkinsonian syndromes and the most frequent neurodegenerative disease after AD. PD is the consequence of the degeneration of dopaminergic neurons in the substantia nigra. The pathological hallmark of PD is the presence of intracytoplasmic inclusions (Lewy bodies) in these neurons. PD cardinal signs include rest tremor, bradykinesia, extrapyramidal rigidity, and postural instability.

Incidence studies have provided estimates ranging between 10 and 15 per 100 000 person years [12]; the lifetime risk of PD is of approximately 1.5% [13]. PD is exceptional before the age of 40; its incidence increases sharply with age and is approximately 1.5 times higher in men than in women at all ages [13]. PD prevalence ranges between 100 and 200 per 100 000 persons overall, and between 1.5% and 2.0% after the age of 65 [14].

There is no cure for the disease and only symptomatic treatments are available; any preventive measure would therefore be useful. Although PD treatment has improved in recent years, PD patients have decreased quality of life [15], and face an increased risk of dementia, institutionalization [16], and death [17].

#### 3.2. Some examples of gene–environment interactions in Parkinson's disease

PD is considered a multifactorial disease resulting from the effect of environmental factors and genetic susceptibility [18]. It is most often sporadic, but approximately 10% to 15% of PD cases have a positive family history of PD among first-degree relatives [19]. Families with autosomal dominant and recessive patterns of inheritance have been described. In recent years, 13 loci have been localized and mutations in seven genes (alpha-synuclein, Parkin, UCH-L1, PINK1, DJ1, LRRK2, Omi) have been identified [20].

Mutations in the LRRK2 gene are responsible for an autosomal form of PD [21]; the penetrance of the most common mutation (G2019S) is incomplete and the lifetime prevalence of PD in mutations carriers has been estimated at 35% [22]. It has been hypothesized that incomplete penetrance in LRRK2 mutation carriers may result from gene–gene or gene–environment interactions, but the interacting genetic or environmental factors have not been identified as yet.

There is also evidence that genetic susceptibility plays a role in the aetiology of sporadic PD. Candidate genes have been associated with PD and can be broadly grouped into four categories: genes involved in the metabolism of xenobiotics (e.g., CYP2D6, NAT2, GSTs), neurodegeneration (e.g., NOS), or the functioning of dopaminergic neurons (e.g., dopamine transporters and receptors), and linkage-derived genes (e.g., UCHL1, alpha-synuclein).

In addition to increasing age and male sex, it is likely that other environmental exposures influence the risk of PD. Whether environmental exposures interact with genetic susceptibility or not is the focus of active research. Our review will focus on two environmental exposures that have been consistently associated with PD, namely cigarette smoking and pesticide exposure, and we will present some examples of gene–environment interactions involving these exposures.

##### 3.2.1. Cigarette smoking and Parkinson's disease

The vast majority of case-control studies that investigated the relation between PD and cigarette smoking have reported an inverse association, which has now

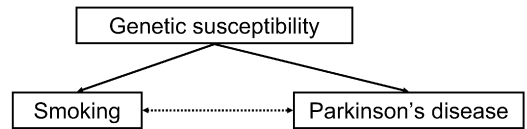
been confirmed in large-cohort studies. A meta-analysis of case-control and cohort studies ( $n = 48$ ) reported an OR of 0.6 (IC 95% = 0.5–0.7) for ever smoking and a dose–effect relation [23].

The origin of this association remains controversial, and several hypotheses have been considered. It has been hypothesized that this relation may result from a true biological protective effect of cigarette smoking and there are experimental data in agreement with this hypothesis: for instance, the mono-amine oxidase B (MAO-B) enzyme, which is involved in oxidative stress, is inhibited in the brain of smokers; stimulation of nicotinic acetylcholine receptors leads to dopamine release; nicotine has a neuroprotective effects for dopaminergic neurons in the nigrostriatal tract [24].

Epidemiologic studies have investigated whether this inverse association may result from bias. Biases that would result either from PD patients quitting smoking more often than controls during the pre-symptomatic phase of the disease (cause–effect bias) or from higher smoking-related mortality among incident PD cases than among controls (incidence–prevalence bias) seem unlikely [17,25,26].

The inverse association between PD and smoking may result from confounding. It has been argued that the inverse association may be the consequence of a pre-morbid personality in PD patients characterized by a lower frequency of behaviours associated with novelty seeking [27]. Some studies performed personality assessments in PD patients, but all were carried out after the disease onset and the results of these tests may be modified by the disease; prospective studies are therefore necessary to investigate this hypothesis [28]. It has also been hypothesized that genetic polymorphisms that influence tolerance to tobacco smoke may also increase the risk of PD and account for this association (Fig. 1A). Studies of candidate genes are underway to test this hypothesis. Recent twin studies have made an interesting contribution to this question. A Swedish twin study compared PD patients who had an unaffected twin to two different control groups: one group of unrelated controls and another one of unaffected co-twins [29]. The case-unrelated controls analysis observed an inverse association for smoking similar to that usually observed (OR = 0.56, 95%CI = 0.40–0.79). The affected twin-unaffected co-twin comparison also found an inverse association (OR = 0.64, 95%CI = 0.37–1.10); analyses restricted to monozygotic twins confirmed this finding. The authors concluded that the inverse association between PD and cigarette smoking is only in part explained by genetic or familial confounding factors (since co-twins are matched on familial factors,

### A- Confounding



### B- Interaction

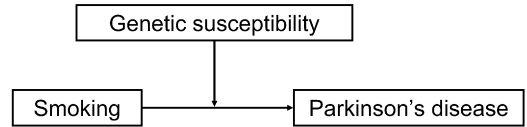


Fig. 1. Hypothetic scenarios involving cigarette smoking, genetic susceptibility, and Parkinson's disease (PD).

and monozygotic twins are matched on genetic background). Another study found that the risk of PD was inversely correlated within twin pairs with the dose of cigarette smoking; this effect was more pronounced in monozygotic twins [30].

In this context, it has been argued that investigating interactions between genes and smoking may contribute to the understanding of the relation between PD and smoking; indeed, showing that cigarette smoking interacts with PD susceptibility genes would suggest that both variables act through a common pathway.

Nitric oxide (NO) is a biological messenger molecule with diverse physiologic roles. However, NO is also a free radical and can combine with superoxide anions to form peroxynitrite. Therefore, NO contributes to oxidative stress and it plays an important role in the aetiology of PD. In brain cells, NO is synthesized by the neuronal (nNOS) and the inducible (iNOS) NO synthases, and inhibition of nNOS or iNOS, either using pharmacological substances or in gene-deficient mice, prevents the destruction of dopaminergic neurons in MPTP models of PD [31]. There is also evidence of NO overproduction as well as of an implication of nNOS and iNOS in PD patients [32]. In a community-based case-control study (TERRE) of PD performed in France among affiliates to the 'Mutualité Sociale Agricole', we found associations between PD and single nucleotide polymorphisms (SNPs) in exon 22 of iNOS (OR for AA carriers = 0.5, 95%CI = 0.3–0.9) and exon 29 of nNOS (OR for carriers of the T allele = 1.5, 95%CI = 1.1–2.2), which suggest that nNOS and iNOS play a role in the susceptibility to sporadic PD [33]. Two further studies investigated the association between iNOS and PD. The inverse association between PD and the exon 22 SNP in iNOS was confirmed in a case-control study per-

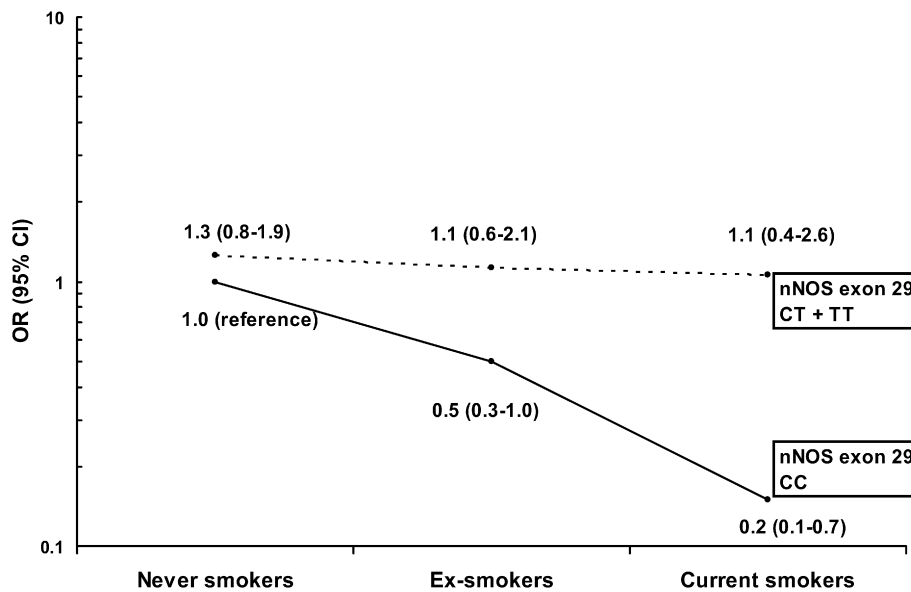


Fig. 2. Interaction between a polymorphism in exon 29 of the nNOS gene and cigarette smoking in Parkinson's disease in the TERRE study (adapted from [36]).

formed in Finland (OR = 0.5, 95%CI = 0.3–0.9) [34]. Another study from the US studied 13 SNPs in the iNOS gene (including the exon 22 SNP) using a family-based design and a statistical method (APL) that allows one to capture information from triads (parents and offspring), discordant siblings, and affected siblings pairs or triads [35]. They found a positive association between PD and the AA genotype of the exon 22 SNP in early-onset families (proband with onset before 40 years); in addition the A allele was also associated with decreased age at onset among all families. Thus, the direction of the association was opposite in this study to that observed in the previous two studies. Numerous differences between the studies could account for this discrepancy; in particular, in the French and Finnish studies, age at onset was considerably higher, and the proportion of familial cases lower compared to the family-based study. In addition, if the exon 22 SNP is not a causal variant, but is in linkage disequilibrium with it, different genetic structures of the populations resulting in different linkage disequilibrium patterns may be responsible for the opposite direction of the associations.

The relation between cigarette smoking and NO remains unclear, but it has been shown that nicotine affects NO production [36] and that cigarette smoke condensates inhibit inflammatory induction of iNOS and reduced cytotoxic effects [37]. We investigated whether SNPs in nNOS and iNOS modified the relation between PD and cigarette smoking (Fig. 1B). As shown in Fig. 2, we observed an interaction ( $p = 0.04$ ) between the exon

29 SNP in the nNOS gene and cigarette smoking; the inverse association between PD and smoking was lost among carriers of the polymorphic T allele, while it was present among CC homozygotes. The relation between smoking and PD was not significantly modified by iNOS; however, because of the low frequency of AA homozygotes, our study was underpowered to detect modest interactions. In the family-based study from the US, the inverse association between PD and smoking was only present among homozygotes for the wild allele of the exon 22 SNP in the iNOS gene (OR = 0.3, 9%CI = 0.1–0.6), and it disappeared among carriers of the A allele (OR = 0.9, 9%CI = 0.6–1.4) ( $p$  for interaction <0.0001) [35].

Altogether, these studies suggest that nNOS and iNOS play a role in PD susceptibility. Although the exact mechanism accounting for the interaction between smoking and the genetic factors remains unclear and merits further studies, it is in favour of a protective effect of cigarette smoking for PD.

### 3.2.2. Farming, pesticide exposure and Parkinson's disease

Twenty years ago, the discovery that MPTP exposure induced Parkinsonian syndromes triggered studies on the relation between pesticide exposure and PD [38]. MPTP is a neurotoxic that is selectively transported into dopaminergic neurons and is metabolized to MPP<sup>+</sup>, a potent mitochondrial complex I inhibitor. MPTP has a

Table 1

The relation between CYP2D6\*4, pesticide exposure, and Parkinson's disease in the TERRE study in France (adapted from [53])

CYP2D6*4	Exposure to pesticides, OR (95%CI), <i>p</i>					
	No exposure		Gardening use		Professional use	
0 or 1 allele	1.00	–	1.3 (0.8–2.4)	0.31	1.7 (0.9–3.0)	0.10
2 alleles (PMs)	0.4 (0.1–3.5)	0.37	2.5 (0.5–12.0)	0.27	4.2 (1.2–15.0)	0.03

ORs (95%CI) and *p*-values were calculated using conditional logistic regression for matched sets and adjusted for ever cigarette smoking.

chemical structure close to that of Paraquat, a widely used herbicide still available in many countries.

Ecologic studies and case-control studies performed in different countries have shown an association between PD and farming or professional pesticide use. A meta-analysis of some of these studies reported ORs of 1.4 (95%CI = 1.1–1.9) for farming and 1.9 (95%CI = 1.5–2.5) for professional pesticide use [39,40]. This relation has been shown to be independent of cigarette smoking [25]. Few studies studied specific product categories or dose–effect relations [41,42]. A recent review of all studies conducted until 2005 concluded that there was stronger evidence for herbicides and insecticides than for fungicides [43].

This relation was only recently studied in a large-cohort study. In the Cancer Prevention Study II Nutrition Cohort, individuals exposed to pesticides had a 70% higher incidence of PD than those not exposed (RR = 1.7, 95% IC = 1.2–2.3); interestingly, this association was similar in farmers and non-farmers and no relation was found between PD and several other chemical exposures (e.g., asbestos, acids, solvents, coal) [44].

The findings from epidemiologic studies have motivated laboratory studies in vitro and in vivo that show that some pesticides are neurotoxic; the detailed commentary of these studies is beyond the scope of this paper and the reader will find a detailed review elsewhere [43].

All the epidemiologic studies raise the issue of how pesticide use was assessed. The retrospective evaluation of pesticide exposure is complex, given the large number of available products and their evolution over time, important variations in methods of application, and the long period of use (with often concomitant exposure to several products). In the majority of the studies, pesticide exposure was assessed using very simple questions and in a dichotomous way, without taking into account timing or importance of exposure. Only two studies used a job-exposure matrix [42,45], and only one (TERRE study) [46] used a detailed in person assessment procedure by occupational health physicians [47].

It has been hypothesized that the relation between pesticides and PD involves enzymes that regulate the disposition of xenobiotics [18,48], including their meta-

bolism and transport. The most extensively studied gene has been the cytochrome P450 2D6 (CYP2D6) that metabolizes environmental toxics, including pesticides (organophosphates, atrazine) and the neurotoxin MPTP. CYP2D6 activity is genetically determined. Poor metabolizers (PMs) have undetectable CYP2D6 activity and represent 5–10% of Caucasians. This trait is inherited as a recessive autosomal trait, and the most frequent polymorphism among Caucasian PMs is a G/A transition at intron 3-exon 4 junction (CYP2D6\*4). Many studies on the relation between CYP2D6 PMs and PD have been performed and a meta-analysis of these studies reported an OR of 1.5 (95%CI = 1.2–2.0) [49]. We performed a study in a population characterized by a high prevalence of professional pesticide exposure in France, in which pesticide exposure was assessed using a two-stage procedure by occupational health physicians [46]. As shown in Table 1, the relation between pesticide exposure and PD was stronger among CYP2D6 PMs than in those who were not, thus suggesting a CYP2D6 by pesticide interaction. A similar interaction was later confirmed by an Australian study [50].

PET studies have shown a brain–blood barrier (BBB) dysfunction in PD patients, likely due to reduced P-glycoprotein (P-gp) function [51]. P-gp actively transports a wide range of molecules from the brain side to the blood side of the blood–brain barrier, and is encoded by the MDR1 gene. Two exonic polymorphisms (e21/2677[G/T/A], e26/3435[C/T]) are associated with differences in MDR1 expression or function. It has been hypothesized that MDR1 decreased function may be involved in the association between PD and pesticides, but previous case-control studies yielded inconsistent findings [52–54]. According to preliminary data from the TERRE study, the relation between PD and specific pesticides that are known to interact with the P-gP protein (organophosphates, organochlorines) is modified by the e21 polymorphism in the MDR1 gene [55]. Thus, pesticides that cross the BBB may be less efficiently transferred to blood in patients with lower P-gP activity. A better understanding of the relation between P-gp and specific pesticides and of the functionality of MDR1 polymorphisms is needed.

The dopamine transporter (DAT) is another transporter and plays a critical role in dopaminergic neurotransmission. In a case-control study from the US, 22 SNPs in the 5' region and a VNTR polymorphism in the 3' region of the DAT gene (SLC6A3) were studied [56]. The 22 polymorphisms segregate as eight haplotypes that fit into two main clades. Having two or more 5' and 3' risk alleles was associated with a modest increase in PD risk (OR = 1.6, 95%CI = 1.1–2.4); the OR increased to 5.7 (96%CI = 1.7–18.5) among occupationally pesticide-exposed subjects. Laboratory studies have shown that organochlorine and pyrethroid insecticides can alter dopaminergic transmission in animals by increasing DAT expression [57,58]. DAT acts as a transporter, but there has been no proof that pesticides enter the neurons through this way. If this assumption is correct, these findings suggest that some pesticides exert their toxic effects to dopaminergic neurons without entering the neuron through DAT [56].

If replicated in other studies, these examples of gene by pesticide interaction in PD may contribute to a better understanding of the mechanisms linking pesticide exposure to PD. If the role of pesticide exposure was confirmed in the aetiology of PD, simple measures, such as improved education and protection during pesticide handling, should be reinforced. The attributable risk of PD related to professional exposure to pesticides is likely to be small and to vary across countries, due to different frequencies and patterns of exposure. However, if it was confirmed that genes interact with pesticides to increase the risk of PD, it is possible that exposures at lower doses observed in non-professional settings may be harmful, thus increasing the attributable risk and raising important issues about the effect of exposure at lower doses.

#### 4. Gene–environment interactions in Alzheimer's disease

The prevalence of dementia in European populations aged 65 and older ranges between 5% and 10%. Dementia prevalence doubles every 4-year increase in age, reaching approximately 30% at age 80. Dementia incidence is about 1% per year in subjects aged 65 and older [59].

Alzheimer's disease (AD) accounts for the vast majority of age-related dementia. AD is an insidious and progressive neurodegenerative disorder that impairs different cognitive functions, resulting in global cognitive dysfunction that interferes with daily life activities. AD is the first cause of institutionalisation in elderly population. Pathologically, the disease is characterized by

accumulation of A $\beta$  protein deposits and neurofibrillary tangles in the brain.

Family history of dementia is the second-greatest risk factor for the disease after age, and the growing understanding of AD genetics has been central to the knowledge of the pathogenic mechanisms leading to the disease.

Genetically, AD is complex and heterogeneous and appears to follow an age-related dichotomy: rare and highly penetrant early-onset familial AD (EOFAD) mutations in different genes are transmitted in an autosomal dominant pattern of inheritance, while late-onset AD (LOAD) without obvious familial segregation is thought to be explained by the 'Common disease/Common variant' hypothesis.

EOFAD represents only a small fraction of all AD cases ( $\leq 5\%$ ) and typically presents with onset ages younger than 65 years. To date, more than 160 mutations in three genes have been reported to cause EOFAD. These include the A $\beta$  precursor protein (APP) on chromosome 21, *presenilin 1* (PSEN1) on chromosome 14, and *presenilin 2* (PSEN2) on chromosome 1 [60]. The most frequently mutated gene, PSEN1, accounts for the majority of AD cases with onset prior to age 50. While these AD-causing mutations occur in three different genes located on three different chromosomes, they all share a common biochemical pathway, i.e., the altered production of A $\beta$  leading to a relative overabundance of the A $\beta$ 42 species, which eventually results in neuronal cell death and dementia.

LOAD, on the other hand, is classically defined as AD with onset at age 65 years or older and represents the vast majority of all AD cases. Segregation and twin studies conclusively suggest a major role of genetic factors in this form of AD [61]. To date, only one genetic factor for LOAD has been established, the  $\epsilon 4$  allele of the *apolipoprotein E* gene (APOE) on chromosome 19q13. The  $\epsilon 4$  allele increases "susceptibility" to AD but is not directly causal. Possession of one  $\epsilon 4$  allele is associated with a 2- to 3-fold increased risk, while having two copies is associated with a 5-fold increase. APOE polymorphism has a major effect on the age at AD clinical onset, which decreases when the number of  $\epsilon 4$  alleles (0, 1, or 2) increases. Because APOE- $\epsilon 4$  is a common allelic variant, the attributable risk associated with  $\epsilon 4$ , at the general population level, has been estimated at 20%, making it the most important risk factor for AD in elderly individuals. Despite this genetic association is long known and well established, the biochemical role of APOE- $\epsilon 4$  in AD pathogenesis is not yet fully understood.

Cohort studies have consistently found that illiteracy, low educational achievement, or a poor socioeconomic environment in early life was a risk factor for late onset of AD. In the same line, occupation involving intellectual activities and time engaged in mental activities in late life may be also associated with decreased risk of AD. Although these associations have been reported by most studies, their interpretation remains controversial. The privileged explanation refers to the concept of ‘cognitive reserve’. According to this hypothesis, persons with high educational or occupational achievement have cognitive reserve that allows compensating, at least for some extent, the cognitive consequences of the neurodegeneration.

No specific environmental risk factor was found to be consistently associated with AD. Studies have reported associations of AD with depression, traumatic head injury, and cardiovascular-related disorders such as hypertension, myocardial infarct, hypercholesterolemia, and stroke. However, it remains unclear whether these are true risk factors or simply co-morbidities that increase severity of cognitive disorders. Use of post-menopausal hormonal treatment, anti-inflammatory drugs and lipid-lowering drugs (statins), regular moderate wine in moderate consumption, and smoking have been found associated with decreased risk of AD in both case-control and cohort studies. Randomized trials have not confirmed the protective effect of hormone replacement therapy on the risk of AD.

Because of the strong association between APOE- $\epsilon 4$  and AD, many of these studies have added stratified analyses looking for interaction between non-genetic risk factors of AD and presence or absence of the  $\epsilon 4$  allele. Results have been inconsistent. In the Epidemiology of Vascular Aging (EVA) study, we found that the inverse association between AD and moderate alcohol consumption was restricted to non-carriers of the  $\epsilon 4$  allele, while moderate alcohol consumption was associated with an increased risk of AD in  $\epsilon 4$  carriers [62]. The Rotterdam Study found a similar interaction between  $\epsilon 4$  and smoking. However, neither the EVA nor the Rotterdam results have been confirmed by other studies. Overall, studies found that the strength of the association between  $\epsilon 4$  and the risk of AD decreased with increasing age, but this can result from complex bias that has been described here above, rather than be due to a biological interaction between  $\epsilon 4$  and an unknown age-dependent factor [62–70].

The apolipoprotein E plays an important role in transport and metabolism of plasma cholesterol. A few studies have shown an interaction between cholesterol, APOE and AD [71–76]. Increased levels of total chole-

sterol were associated with an increased risk of AD only in individuals without the  $\epsilon 4$  allele [72,75,76]. But other studies have failed to show any interaction [74]. In any case, the biological mechanism underlying this potential interaction remains unclear.

## 5. Conclusions

In recent years, genetic studies in PD have provided interesting lessons. By focusing on candidate genes with strong biological plausibility (e.g., alpha-synuclein), evidence for the role of genetic susceptibility in sporadic PD has been obtained [77]. Collaborative studies including large numbers of affected subjects have started to be performed and yielded important findings [77,78]. Studies investigating the interaction between genetic susceptibility and environmental exposures have been performed, and while their results remain to be replicated, they show the interest of focusing on genes with known functional consequences and mechanisms with a priori biological plausibility. The situation is very different in sporadic AD. A single gene has been identified (apolipoprotein E), which explains a large part of the variability of late-onset AD risk. Few environmental risk factors have been consistently associated with AD. Some studies have systematically investigated interactions between the APOE polymorphism and possible non-genetic factors of AD risk. The few interactions that have been described had no biological plausibility, and they can be considered as chance results.

## References

- [1] C.J.L. Murray, A.D. Lopez, The Global Burden of Disease, World Health Organisation, Geneva, 1996.
- [2] M. Menken, T.L. Munsat, J.F. Toole, The global burden of disease study: implications for neurology, *Arch. Neurol.* 57 (2000) 418–420.
- [3] L.I. Golbe, Neurodegeneration in the age of molecular biology, *BMJ* 324 (2002) 1467–1468.
- [4] J.P. Ioannidis, T.A. Trikalinos, M.J. Khoury, Implications of small effect sizes of individual genetic variants on the design and interpretation of genetic association studies of complex diseases, *Am. J. Epidemiol.* 164 (2006) 609–614.
- [5] M.Y. Wong, N.E. Day, J.A. Luan, K.P. Chan, N.J. Wareham, The detection of gene–environment interaction for continuous traits: should we deal with measurement error by bigger studies or better measurement? *Int. J. Epidemiol.* 32 (2003) 51–57.
- [6] M. Garcia-Closas, W.D. Thompson, J.M. Robins, Differential misclassification and the assessment of gene–environment interactions in case-control studies, *Am. J. Epidemiol.* 147 (1998) 426–433.
- [7] A. Schatzkin, E. Slud, Competing risks bias arising from an omitted risk factor, *Am. J. Epidemiol.* 129 (1989) 850–856.
- [8] E. Slud, D. Byar, How dependent causes of death can make risk factors appear protective, *Biometrics* 44 (1988) 265–269.



- [9] A. Elbaz, A. Alperovitch, Bias in association studies resulting from gene–environment interactions and competing risks, *Am. J. Epidemiol.* 155 (2002) 265–273.
- [10] L.A. Farrer, L.A. Cupples, J.L. Haines, B. Hyman, W.A. Kukull, R. Mayeux, R.H. Myers, M.A. Pericak-Vance, N. Risch, C.M. Van Duijn, Effects of age, sex, and ethnicity on the association between apolipoprotein E genotype and Alzheimer disease. A meta-analysis. APOE and Alzheimer Disease Meta Analysis Consortium, *J. Am. Med. Assoc.* 278 (1997) 1349–1356.
- [11] W.A. Rocca, S.K. McDonnell, K.J. Strain, J.H. Bower, J.E. Ahlskog, A. Elbaz, D.J. Schaid, D.M. Maraganore, Familial aggregation of Parkinson's disease: The Mayo Clinic family study, *Ann. Neurol.* 56 (2004) 495–502.
- [12] D. Twelves, K.S.M. Perkins, C. Counsell, Systematic review of incidence studies of Parkinson's disease, *Mov. Disord.* 18 (2003) 19–31.
- [13] A. Elbaz, J.H. Bower, D.M. Maraganore, S.K. McDonnell, B.J. Peterson, J.E. Ahlskog, D.J. Schaid, W.A. Rocca, Risk tables for parkinsonism and Parkinson's disease, *J. Clin. Epidemiol.* 55 (2002) 25–31.
- [14] M.C. de Rijk, L.J. Launer, K. Berger, M.M. Breteler, J.F. Dartigues, M. Baldereschi, L. Fratiglioni, A. Lobo, J. Martinez-Lage, C. Trenkwalder, A. Hofman, Prevalence of Parkinson's disease in Europe: A collaborative study of population-based cohorts. Neurologic Diseases in the Elderly Research Group, *Neurology* 54 (2000) S21–S23.
- [15] P. Martinez-Martin, An introduction to the concept of 'quality of life in Parkinson's disease', *J. Neurol.* 245 (1998) S2–S6.
- [16] S.A. Parashos, D.M. Maraganore, P.C. O'Brien, W.A. Rocca, Medical services utilization and prognosis in Parkinson disease: a population-based study, *Mayo Clin. Proc.* 77 (2002) 918–925.
- [17] A. Elbaz, J.H. Bower, B.J. Peterson, D. Maraganore, S.K. McDonnell, J.E. Ahlskog, D.J. Schaid, W.A. Rocca, Survival study of Parkinson Disease in Olmsted County, Minnesota, *Arch. Neurol.* 60 (2003) 91–96.
- [18] H. Checkoway, L.M. Nelson, Epidemiologic approaches to the study of Parkinson's disease etiology, *Epidemiology* 10 (1999) 327–336.
- [19] A. Elbaz, S.K. McDonnell, D.M. Maraganore, K.J. Strain, D.J. Schaid, J.H. Bower, J.E. Ahlskog, W.A. Rocca, Validity of family history data on PD: Evidence for a family information bias, *Neurology* 61 (2003) 11–17.
- [20] M.J. Farrer, Genetics of Parkinson disease: paradigm shifts and future prospects, *Nat. Rev. Genet.* 7 (2006) 306–318.
- [21] A. Zimprich, S. Biskup, P. Leitner, P. Lichtner, M. Farrer, S. Lincoln, J. Kachergus, M. Hulihan, R.J. Uitti, D.B. Calne, A.J. Stoessl, R.F. Pfeiffer, N. Patenge, I.C. Carbajal, P. Vieregge, F. Asmus, B. Muller-Mysok, D.W. Dickson, T. Meitinger, T.M. Strom, Z.K. Wszolek, T. Gasser, Mutations in LRRK2 cause autosomal-dominant parkinsonism with pleomorphic pathology, *Neuron* 44 (2004) 601–607.
- [22] L.J. Ozelius, G. Senthil, R. Saunders-Pullman, E. Ohmann, A. Deligtisch, M. Tagliati, A.L. Hunt, C. Klein, B. Henick, S.M. Hailpern, R.B. Lipton, J. Soto-Valencia, N. Risch, S.B. Bressman, LRRK2 G2019S as a cause of Parkinson's disease in Ashkenazi Jews, *N. Engl. J. Med.* 354 (2006) 424–425.
- [23] M.A. Hernan, B. Takkouche, F. Caamanolsorna, J.J. Gestal-Otero, A meta-analysis of coffee drinking, cigarette smoking, and the risk of Parkinson's disease, *Ann. Neurol.* 52 (2002) 276–284.
- [24] G.W. Ross, H. Petrovitch, Current evidence for neuroprotective effects of nicotine and caffeine against Parkinson's disease, *Drug Aging* 18 (2001) 797–806.
- [25] J.P. Galanaud, A. Elbaz, J. Clavel, J.-S. Vidal, J.-R. Corrèze, A. Alperovitch, C. Tzourio, Cigarette smoking and Parkinson's disease: a case-control study in a population characterized by a high prevalence of pesticide exposure, *Mov. Disord.* 20 (2005) 181–189.
- [26] H. Chen, S.M. Zhang, M.A. Schwarzschild, M.A. Hernan, A. Ascherio, Survival of Parkinson's disease patients in a large prospective cohort of male health professionals, *Mov. Disord.* 21 (2006) 1002–1007.
- [27] M.A. Menza, L.I. Golbe, R.A. Cody, N.E. Forman, Dopamine-related personality traits in Parkinson's disease, *Neurology* 43 (1993) 505–508.
- [28] L. Ishihara, C. Brayne, What is the evidence for a premorbid parkinsonian personality: A systematic review, *Mov. Disord.* 21 (2006) 1066–1072.
- [29] K. Wirdefeldt, M. Gatz, Y. Pawitan, N.L. Pedersen, Risk and protective factors for Parkinson's disease: a study in Swedish twins, *Ann. Neurol.* 57 (2005) 27–33.
- [30] C.M. Tanner, S.M. Goldman, D.A. Aston, R. Ottman, J. Ellenberg, R. Mayeux, J.W. Langston, Smoking and Parkinson's disease in twins, *Neurology* 58 (2002) 581–588.
- [31] P. Hantraye, E. Brouillet, R. Ferrante, S. Palfi, R. Dolan, R.T. Matthews, M.F. Beal, Inhibition of neuronal nitric oxide synthase prevents MPTP-induced parkinsonism in baboons, *Nat. Med.* 2 (1996) 1017–1021.
- [32] S. Hunot, F. Boissiere, B. Faucheux, B. Brugg, A. Mouatt-Prigent, Y. Agid, E.C. Hirsch, Nitric oxide synthase and neuronal vulnerability in Parkinson's disease, *Neuroscience* 72 (1996) 355–363.
- [33] C. Levecque, A. Elbaz, J. Clavel, F. Richard, J.-S. Vidal, P. Amouyel, C. Tzourio, A. Alperovitch, M.-C. Chartier-Harlin, Association between Parkinson's disease and polymorphisms in the nNOS and iNOS genes in a community-based case-control study, *Hum. Mol. Genet.* 12 (2003) 79–86.
- [34] S. Hague, T. Peuralinna, J. Eerola, O. Hellstrom, P.J. Tienari, A.B. Singleton, Confirmation of the protective effect of iNOS in an independent cohort of Parkinson disease, *Neurology* 62 (2004) 635–636.
- [35] D.B. Hancock, E.R. Martin, K. Fujiwara, M.A. Stacy, B.L. Scott, J.M. Stajich, R. Jewett, Y.J. Li, M.A. Hauser, J.M. Vance, W.K. Scott, NOS2A and the modulating effect of cigarette smoking in Parkinson's disease, *Ann. Neurol.* 60 (2006) 366–373.
- [36] S. Pogun, S. Demigoren, D. Taskiran, L. Kanit, O. Yilmaz, E.O. Koylu, B. Balkan, E.D. London, Nicotine modulates nitric oxide in rat brain, *Eur. Neuropsychopharmacol.* 10 (2000) 463–472.
- [37] E.A. Mazzio, M.G. Kolta, R.R. Reams, K.F. Soliman, Inhibitory effects of cigarette smoke on glial inducible nitric oxide synthase and lack of protective properties against oxidative neurotoxins in vitro, *Neurotoxicology* 26 (2005) 49–62.
- [38] J.W. Langston, P. Ballard, J.W. Tetrud, I. Irwin, Chronic Parkinsonism in humans due to a product of meperidine-analog synthesis, *Science* 219 (1983) 979–980.
- [39] A. Priyadarshi, S.A. Khuder, E.A. Schaub, S. Shrivastava, A meta-analysis of Parkinson's disease and exposure to pesticides, *Neurotoxicology* 21 (2000) 435–440.
- [40] A. Priyadarshi, S.A. Khuder, E.A. Schaub, S.S. Priyadarshi, Environmental risk factors and Parkinson's disease: a metaanalysis, *Environ. Res.* 86 (2001) 122–127.
- [41] H.H. Liou, M.C. Tsai, C.J. Chen, J.S. Jeng, Y.C. Chang, S.Y. Chen, R.C. Chen, Environmental risk factors and Parkinson's

- disease: A case-control study in Taiwan, *Neurology* 48 (1997) 1583–1588.
- [42] A. Seidler, W. Hellenbrand, B.P. Robra, P. Vieregge, P. Nischan, J. Joerg, W.H. Oertel, G. Ulm, E. Schneider, Possible environmental, occupational, and other etiologic factors for Parkinson's disease: a case-control study in Germany, *Neurology* 46 (1996) 1275–1284.
- [43] T.P. Brown, P.C. Rumsby, A.C. Capleton, L. Rushton, L.S. Levy, Pesticides and Parkinson's disease – is there a link? *Environ. Health Perspect.* 114 (2006) 156–164.
- [44] A. Ascherio, H. Chen, M.G. Weisskopf, E.J. O'Reilly, M.L. McCullough, E.E. Calle, M.A. Schwarzschild, M.J. Thun, Pesticide exposure and Parkinson's disease, *Ann. Neurol.* 60 (2006) 197–203.
- [45] I. Baldi, P. Lebaillly, B. Mohammed-Brahim, L. Letenneur, J.-F. Dartigues, P. Brochard, Neurodegenerative diseases and exposure to pesticides in the elderly, *Am. J. Epidemiol.* 157 (2003) 409–414.
- [46] A. Elbaz, C. Levecque, J. Clavel, J.-S. Vidal, F. Richard, P. Amouyel, A. Alperovitch, M.C. Chartier-Harlin, C. Tzourio, CYP2D6 polymorphism, pesticides exposure, and Parkinson's disease, *Ann. Neurol.* 55 (2004) 430–434.
- [47] W.F. Stewart, P.A. Stewart, Occupational case-control studies: I. Collecting information on work histories and work-related exposures, *Am. J. Ind. Med.* 26 (1994) 297–312.
- [48] D.G. LeCouteur, A.J. McLean, M.C. Taylor, B.L. Woodham, P.G. Board, Pesticides and Parkinson's disease, *Biomed. Pharmacother.* 53 (1999) 122–130.
- [49] S.J. McCann, S.M. Pond, K.M. James, D.G. LeCouteur, The association between polymorphisms in the cytochrome P-450 2D6 gene and Parkinson's disease: a case-control study and meta-analysis, *J. Neurol. Sci.* 153 (1997) 50–53.
- [50] Y. Deng, B. Newman, M.P. Dunne, P.A. Silburn, G.D. Mellick, Further evidence that interactions between CYP2D6 and pesticide exposure increase risk for Parkinson's disease, *Ann. Neurol.* 55 (2004) 897.
- [51] R. Kortekaas, K.L. Leenders, J.C. van Oostrom, W. Vaalburg, J. Bart, A.T. Willemsen, N.H. Hendrikse, Blood-brain barrier dysfunction in parkinsonian midbrain in vivo, *Ann. Neurol.* 57 (2005) 176–179.
- [52] E.K. Tan, D.K. Chan, P.W. Ng, J. Woo, Y.Y. Teo, K. Tang, L.P. Wong, S.S. Chong, C. Tan, H. Shen, Y. Zhao, C.G. Lee, Effect of MDR1 haplotype on risk of Parkinson disease, *Arch. Neurol.* 62 (2005) 460–464.
- [53] T. Furuno, M.T. Landi, M. Ceroni, N. Caporaso, I. Bernucci, G. Nappi, E. Martignoni, E. Schaeffeler, M. Eichelbaum, M. Schwab, U.M. Zanger, Expression polymorphism of the blood-brain barrier component P-glycoprotein (MDR1) in relation to Parkinson's disease, *Pharmacogenetics* 12 (2002) 529–534.
- [54] M. Drozdziak, M. Bialecka, K. Mysliwiec, K. Honczarenko, J. Stankiewicz, Z. Sych, Polymorphism in the P-glycoprotein drug transporter MDR1 gene: a possible link between environmental and genetic factors in Parkinson's disease, *Pharmacogenetics* 13 (2003) 259–263.
- [55] A. Elbaz, F. Duthel, A. Alperovitch, M.-A. Liorot, C. Tzourio, Case-control study of the MDR1 gene in Parkinson disease, *Mov. Disord.* 21 (15) (2006) S405 (suppl.).
- [56] S.N. Kelada, H. Checkoway, S.L. Kardia, C.S. Carlson, P. Costamallen, D.L. Eaton, J. Firestone, K.M. Powers, P.D. Swanson, G.M. Franklin, W.T. Longstreth Jr., T.S. Weller, Z. Afsharinejad, L.G. Costa, 5' and 3' region variability in the dopamine transporter gene (SLC6A3), pesticide exposure and Parkinson's disease risk: a hypothesis-generating study, *Hum. Mol. Genet.* 15 (2006) 3055–3062.
- [57] J.R. Richardson, W.M. Caudle, M. Wang, E.D. Dean, K.D. Pennell, G.W. Miller, Developmental exposure to the pesticide diieldrin alters the dopamine system and increases neurotoxicity in an animal model of Parkinson's disease, *FASEB J.* 20 (2006) 1695–1697.
- [58] W.M. Caudle, J.R. Richardson, M. Wang, G.W. Miller, Perinatal heptachlor exposure increases expression of presynaptic dopaminergic markers in mouse striatum, *Neurotoxicology* 26 (2005) 721–728.
- [59] K. Ritchie, S. Lovestone, The dementias, *Lancet* 360 (2002) 1759–1766.
- [60] R. Mayeux, Genetic epidemiology of Alzheimer disease, *Alzheimer Dis. Assoc. Disord.* 20 (2006) S58–S62.
- [61] R. Mayeux, M. Sano, J. Chen, T. Tatemichi, Y. Stern, Risk of dementia in first-degree relatives of patients with Alzheimer's disease and related disorders, *Arch. Neurol.* 48 (1991) 269–273.
- [62] C. Dufouil, C. Tzourio, C. Brayne, C. Berr, P. Amouyel, A. Alperovitch, Influence of apolipoprotein E genotype on the risk of cognitive deterioration in moderate drinkers and smokers, *Epidemiology* 11 (2000) 280–284.
- [63] M.N. Haan, L. Shemanski, W.J. Jagust, T.A. Manolio, L. Kuller, The role of APOE epsilon 4 in modulating effects of other risk factors for cognitive decline in elderly persons, *J. Am. Med. Assoc.* 282 (1999) 40–46.
- [64] L. Zhu, L. Fratiglioni, Z.C. Guo, H. Basun, E.H. Corder, B. Winblad, M. Viitanen, Incidence of dementia in relation to stroke and the apolipoprotein E epsilon 4 allele in the very old – Findings from a population-based longitudinal study, *Stroke* 31 (2000) 53–60.
- [65] K. Yaffe, M. Haan, A. Byers, C. Tangen, L. Kuller, Estrogen use, APOE, and cognitive decline – Evidence of gene–environment interaction, *Neurology* 54 (2000) 1949–1953.
- [66] R. Peila, L.R. White, H. Petrovich, K. Masaki, G.W. Ross, R.J. Havlik, L.J. Launer, Joint effect of the APOE gene and midlife systolic blood pressure on late-life cognitive impairment – The Honolulu–Asia aging study, *Stroke* 32 (2001) 2882–2887.
- [67] A. Ruitenberg, J.C. vanSwieten, J.C.M. Witteman, K.M. Mehta, C.M. vanDuijn, A. Hofman, M.M.B. Breteler, Alcohol consumption and risk of dementia: the Rotterdam Study, *Lancet* 359 (2002) 281–286.
- [68] R. Peila, B.L. Rodriguez, L.J. Launer, Type 2 diabetes, APOE gene, and the risk for dementia and related pathologies – The Honolulu–Asia Aging Study, *Diabetes* 51 (2002) 1256–1262.
- [69] A. Hofman, A. Ott, M.M.B. Breteler, M.L. Bots, A.J.C. Slooter, F. vanHarskamp, C.M. vanDuijn, C. VanBroeckhoven, D.E. Grobbee, Atherosclerosis, apolipoprotein E, and prevalence of dementia and Alzheimer's disease in the Rotterdam Study, *Lancet* 349 (1997) 151–154.
- [70] L.J. Podewils, E. Guallar, L.H. Kuller, L.P. Fried, O.L. Lopez, M. Carlson, C.G. Lyketsos, Physical activity, APOE genotype, and dementia risk: findings from the Cardiovascular Health Cognition Study, *Am. J. Epidemiol.* 161 (2005) 639–651.
- [71] G.P. Jarvik, E. Wijsman, W.A. Kukull, G.D. Schellenberg, C. Yu, E.B. Larson, Interactions of apolipoprotein E genotype, total cholesterol level, age, and sex in prediction of Alzheimer's disease: a case-control study, *Neurology* 45 (1995) 1092–1096.
- [72] C. Dufouil, F. Richard, N. Fievet, J.F. Dartigues, K. Ritchie, C. Tzourio, P. Amouyel, A. Alperovitch, APOE genotype, cholesterol level, lipid-lowering treatment, and dementia: The Three-City Study, *Neurology* 64 (2005) 1531–1538.

- [73] V. Chandra, R. Pandav, Gene–environment interaction in Alzheimer’s disease: A potential role for cholesterol, *Neuroepidemiology* 17 (1998) 225–232.
- [74] M. Kivipelto, E.L. Helkala, M.P. Laakso, T. Hanninen, M. Hallikainen, K. Alhainen, S. Iivonen, A. Mannermaa, J. Tuomilehto, A. Nissinen, H. Soininen, Apolipoprotein E epsilon 4 allele, elevated midlife total cholesterol level, and high midlife systolic blood pressure are independent risk factors for late-life Alzheimer disease, *Ann. Intern. Med.* 137 (2002) 149–155.
- [75] K. Hall, J. Murrell, A. Ogunniyi, M. Deeg, O. Baiyewu, S. Gao, O. Gureje, J. Dickens, R. Evans, V. Smith-Gamble, F.W. Unverzagt, J. Shen, H. Hendrie, Cholesterol, APOE genotype, and Alzheimer disease: an epidemiologic study of Nigerian Yoruba, *Neurology* 66 (2006) 223–227.
- [76] R.M. Evans, C.L. Emsley, S. Gao, A. Sahota, K.S. Hall, M.R. Farlow, H. Hendrie, Serum cholesterol, APOE genotype, and the risk of Alzheimer’s disease: A population-based study of African Americans, *Neurology* 54 (2000) 240–242.
- [77] D.M. Maraganore, M. De Andrade, A. Elbaz, M.J. Farrer, J.P. Ioannidis, R. Kruger, W.A. Rocca, N. Schneider, T.G. Lesnick, S.J. Lincoln, M.M. Hulihan, J.O. Aasly, T. Ashizawa, M.C. Chartier-Harlin, H. Checkoway, C. Ferrarese, G. Hadjigeorgiou, N. Hattori, H. Kawakami, J.C. Lambert, T. Lynch, G.D. Mellick, S. Papapetropoulos, A. Parsian, A. Quattrone, O. Riess, E.K. Tan, C. van Broeckhoven, Collaborative analysis of the alpha-synuclein gene promoter variability and Parkinson’s disease, *J. Am. Med. Assoc.* 296 (2006) 661–670.
- [78] A. Elbaz, L.M. Nelson, H. Payami, J.P. Ioannidis, B.K. Fiske, G. Annesi, A. Carmine-Belin, S.A. Factor, C. Ferrarese, G. Hadjigeorgiou, D.S. Higgins, H. Kawakami, R. Kruger, K.S. Marder, R.P. Mayeux, G.D. Mellick, J.D. Nutt, B. Ritz, A. Samii, C.M. Tanner, C. van Broeckhoven, S.K. Van Den Eeden, K. Wirdefeldt, C.P. Zabetian, M. Dehem, J.S. Montimurro, A. Southwick, R.M. Myers, T.A. Trikalinos, Lack of replication of thirteen single nucleotide polymorphisms implicated in Parkinson’s disease: a large-scale international study, *Lancet Neurol.* 5 (2006) 917–923.