ABSTRACT: Alzheimer’s disease (AD) is a major problem of health in developed countries and the most important cause of disability in the elderly. More than 200 different genes distributed across the human genome are involved in the pathogenesis of AD. Genetic factors and environmental factors interact to elicit premature neuronal death in dementia. Approximately, 30 percent of the elderly population with dementia suffers both nutritional dysfunction and metabolic disorders potentially contributing to aggravating neuronal deficit and cognitive decline. Some genes associated with pharmacogenomic responses also influence nutrigenomics in AD. Novel nutraceuticals represent a nutrigenomic alternative for a personalized medicine in dementia and cerebrovascular disorders.

INTRODUCTION

Alzheimer’s disease (AD) is the principal type of dementia (40-60 percent), followed by vascular dementia (30-40 percent) and mixed dementia (10-20 percent). More than 25 million people suffer dementia worldwide, and this number may increase to up to 75 million in the coming 20-30 years. The prevalence of AD increases exponentially from approximately 1 percent at 60-65 years of age to more than 30-35 percent in people older than 80 years. Main risk factors for dementia include genetic factors, age, cerebrovascular disorders, and environmental factors (1, 2). During the past 20 years only 5 drugs, 4 cholinesterase inhibitors (tacrine, donepezil, rivastigmine, galantamine) and one NMDA receptor partial antagonist (memantine), with poor cost-effectiveness, have been approved for the treatment of AD. The average annual cost per person with dementia ranges from US$ 15,000 to US$ 50,000, depending upon disease stage and country, with a lifetime cost per patient of more than US$ 175,000. About 10-20 percent of the costs in dementia are attributed to pharmacological treatment, including anti-dementia drugs, psychotropics (antidepressants, neuroleptics, anxiolytics), and other drugs currently prescribed in the elderly. Functional genomics studies have revealed the association of specific mutations in primary loci (APP, PS1, PS2) and/or APOE-related polymorphic variants with the phenotypic expression of biological traits (age at onset, brain atrophy, cognitive decline rate, β-amyloid deposition, lipid metabolism dysfunction, immunological dysregulation, therapeutic outcome). In most pharmacogenomics studies, patients harbouring the APOE-4 allele (especially homozygotes) are the worst responders. Genetic clusters integrating 3-4 AD-related genes, accounting for 25-30 percent of the AD population, have allowed the identification of selective genotype clusters of good responders. Furthermore, approximately 15 percent of the European population with AD shows mutant CYP2D6 alleles (poor and ultrarapid metabolizers) potentially responsible for efficacy and safety problems with cholinesterase inhibitors and psychotropic drugs. Nutritional factors may also contribute to deteriorate cognition and brain function in dementia. Some genes potentially involved in nutrigenetics may also participate in lipid metabolism, brain function, and longevity.

Novel nutraceutical products obtained from marine sources with biotechnological procedures have demonstrated atheroprotective properties and lipid-lowering effects, devoid of hepatotoxic activity. Some of these nutraceuticals exhibit a genotype-dependent therapeutic effect, reflecting a nutrigenomic profile.

GENETICS OF ALZHEIMER’S DISEASE

The genetic defects identified in AD during the past 25 years can be classified into 3 main categories: (a) Mendelian or mutational defects in genes directly linked to AD, including (i) 32 mutations in the amyloid beta (Aβ)(ABP) precursor protein (APP) gene (21q21); (ii) 165 mutations in the presenilin 1 (PS1) gene (14q24.3); and (iii) 12 mutations in the presenilin 2 (PS2) gene (1q31-q42). (b) Multiple polymorphic variants of risk characterized in more than 200 different genes distributed across the human genome can increase neuronal vulnerability to premature death. Among these genes of susceptibility, the apolipoprotein E (APOE) gene (19q13.2) is the most prevalent as a risk factor for AD, especially in those subjects harbouring the APOE-4 allele, whereas carriers of the APOE-2 allele might be protected against dementia. (c) Diverse mutations located in mitochondrial DNA (mtDNA) through heteroplastic transmission can influence aging and oxidative stress conditions, conferring phenotypic heterogeneity. It is also likely that defective functions of genes associated with longevity may influence premature neuronal survival, since neurons are potential pacemakers defining life span in mammals. All these genetic factors may interact in still unknown genetic networks leading to a cascade of pathogenic events characterized by abnormal protein processing and misfolding (conformational changes) with subsequent accumulation of abnormal proteins (protein aggregation), ubiquitin-proteasome system dysfunction, excitotoxic reactions, oxidative and nitrosative stress, mitochondrial injury, synaptic failure, altered metal homeostasis, dysfunction of axonal and dendritic transport, and chaperone misoperation. Some of these mechanisms are common to several neurodegenerative disorders which differ depending upon the gene(s) affected and the involvement of specific genetic networks, together with
cerebrovascular factors, epigenetic factors (DNA methylation) and environmental conditions (nutrition, toxicity, social factors, etc) (3).

**PHARMACOGENOMICs IN DEMENTIA**

It is very likely that the dynamic equilibrium of the mammalian genome resulting from the interaction of constitutional, evolutionary factors (natural selection, positive mutations, polymorphic variants, epigenetic phenomena, genetic epistasis), environmental factors and nutrition, is responsible for the condition of health and well-being, whereas a disequilibrium among these factors might lead to disease conditions. The binomial health-disease conundrum is the consequence of complex interactions between the genome and exogenous factors (nature-nurture functional equilibrium/disruption). The emergence of disease can be managed with drugs that are fully or partially able to restore the equilibrium lost. To understand, from a genomic point of view, how drugs can reconstruct biochemical harmony in the interior milieu, we have to incorporate to our scientific background the tools provided by pharmacogenetics, pharmacogenomics, and nutrigenomics. With pharmacogenetics we can understand how genomic factors associated with genes encoding enzymes responsible for drug metabolism regulate pharmacokinetics and pharmacodynamics (mostly safety issues). With pharmacogenomics we can differentiate the specific disease-modifying effects of drugs (efficacy issues) acting on pathogenic mechanisms directly linked to genes whose mutations determine alterations in protein synthesis or subsequent protein misfolding and aggregation (4-6). The capacity of drugs to reverse the effects of the activation of pathogenic cascades (phenotype expression) regulated by networking genes basically deals with efficacy issues. Finally, with nutrigenomics we can evaluate the effects of the most influential environmental factor (nutrition) on genomic function, disease induction, and drug metabolism, as well as the influence of structural and functional genomics on the appropriate metabolism of nutritional factors (4, 5, 7). It is estimated that genetics accounts for 20 to 95 percent of variability in drug disposition and pharmacodynamics. Recent studies indicate that the therapeutic response in AD is genotype-specific depending upon genes associated with AD pathogenesis and/or genes responsible for drug metabolism (CYPs). In monogenic-related studies, APOE-4/4 carriers are the worst responders. APP, PS1, and PS2 mutations may drastically modify the therapeutic response to conventional drugs. In trigenic (APOE-PS1-PS2 clusters)-related studies the best responders are those patients carrying the 331222-, 341122-, 341222-, and 441112-genomic profiles. The worst responders in all genomic clusters are patients with the 441122-genotype, indicating the powerful, deleterious effect of the APOE-4/4 genotype on therapeutics in networking activity with other AD-related genes (3-7). Cholinesterase inhibitors of current use in AD are metabolized via CYP-related enzymes. These drugs can interact with many other drugs and foods which are substrates, inhibitors or inducers of the cytochrome P-450 system, this interaction eliciting liver toxicity and other adverse drug reactions. CYP2D6-related enzymes are involved in the metabolism of more than 20 percent of CNS drugs. The distribution of the CYP2D6 genotypes differentiates 4 major categories of CYP2D6-related metabolizer types: (i) Extensive Metabolizers (EM) (51.61 percent); (ii) Intermediate Metabolizers (IM)(32.26 percent); (iii) Poor Metabolizers (PM)(9.03 percent); and (iv) Ultra-rapid Metabolizers (UM)(7.10 percent). PMs and UMs tend to show higher transaminase activity than EMs and IMs. EMs and IMs are the best responders, and PMs and UMs are the worst responders to pharmacological treatments in AD. In addition, APOE and CYP2D6 variants interact to define the phenotypic profile of EM, IM, PM and UM (8-12).

**NUTRIGENOMICs IN DEMENTIA**

Nutrigenetics and nutrigenomics are multidisciplinary fields that focus on studying the interactions between nutritional factors, genetic factors and health outcomes. Nutrigenomics refers to the complex effects of the nutritional environment on the genome, epigenome, and proteome, as well as the influence that a particular genomic profile exerts on the metabolic effects of nutritional factors (13, 14). Diet affects gene expression patterns, chromatin organization, and protein post-translational modifications (15). Long-term effects of diet may influence the metabolism of lipids, carbohydrates, proteins, water, major minerals, and trace elements leading to multiple nutrition-related pathologies (i.e., obesity, diabetes, hyperlipidemia, anemia, vitamin deficiencies, hypertension, cardiovascular disorders, metabolic syndrome, cancer, etc), as potential risk factors for cerebrovascular disorders or vascular dementia and aggravating factors for patients with genetic predisposition to suffer AD. Alterations in cholesterol and lipid metabolism may specifically contribute to cerebrovascular disorders and dementia (4, 16). The ‘Genome Health Nutrigenomics’ concept has been introduced to define and focus attention on the specialized research area of how diet impacts on genome stability and how genotype determines nutritional requirements for genome health maintenance (17). There is increasing evidence that risk for degenerative disease increases with more DNA damage, which in turn is dependent on nutritional status, and that the optimal concentration of micronutrients for prevention of genome damage is also dependent on genetic polymorphisms that alter the function of genes involved in the uptake and metabolism of nutrients required for DNA repair and DNA replication (18). Eventually, nutrigenomics will lead to evidence-based dietary intervention strategies for restoring health and fitness and for preventing diet-related disease (19, 20). The importance of nutrition in AD relies on at least 6 different basic aspects: (i) Psychomotor dysfunction, apraxia, dysphagia and behavioural changes in nutritional habits, together with weight loss, may become an important issue in AD daily management with clinical consequences for patients, a greater psychological burden for caregivers, and increased costs for society. (ii) Chronic nutritional deficiency due to either endogenous or exogenous factors may contribute to metabolic dysfunction with direct or indirect repercussions on brain metabolism potentially leading to neurodegeneration. (iii) Specific nutrients and/or essential metabolic factors may be involved in the pathogenic cascades responsible for AD-related premature neurodegeneration. (iv) Different types of food may interfere or facilitate the absorption and gastrointestinal processing of many drugs currently taken by AD patients. (v) AD is a heterogenic, multifactorial disorder in which more than 200 genes in conjunction with diverse environmental factors may affect neuronal survival, contributing to neuronal dysregulation and cell death. (vi) Nutrigenetics/nutrigenomics and pharmacogenetics/pharmacogenomics studies indicate that both nutrients and drugs operate according to a genotype-dependent program in AD (4, 5, 7). Some examples of genes relevant for both nutrigenomics and dementia are methylene tetrahydrofolate reductase (MTHFR), Apolipoprotein A (APOE) and APOA1, leptin, interleukin-1 (IL1), tumour necrosis factor (TNF) and...
peroxisome proliferator-activated receptors (PPARs) (1, 4, 5, 21-24). There are 3 major conceptual aspects related to nutrient-gene interactions with potential influence on aging and dementia: (a) direct interactions in which nutrients behave as transcription factors that can bind to DNA inducing gene expression; (b) epigenetic interactions in which nutrients alter the structure of DNA or histones altering gene expression; and (c) genetic variations by which different SNPs modify or alter gene expression (20). A human being at the age of 80 years has introduced through his/her mouth more than 100-120 tons of food. Both oxygen and daily nutrients are the two most important environmental factors essential for life in mammals. There is a bidirectional influence of nutrients on genomic factors and of genomic factors on the metabolism of nutritional factors to keep a homeostatic equilibrium in health conditions. In addition, many dietary factors may exert a deleterious or toxic effect on neurons (i.e., dietary mutagens, genotoxins, Maillard reaction compounds and melanoidin structures, plant neurotoxins, heavy metals, and ultratrace elements such as aluminium and arsenic), whereas malnutrition or overnutrition may contribute to induce cognitive deterioration either directly or indirectly through concomitant pathologies that affect brain function (7). Major risk factors for cerebrovascular disorders and stroke (prevalence: 1-3 percent) include heart disease (atrial fibrillation, cardiac infarction)(Prevalence: 8-10 percent), lipid metabolism dysfunction (dyslipidemias, hypercholesterolemia, hypertriglyceridemia)(Prevalence: 50-60 percent), obesity (Prevalence: 65 percent with BMI>25.0 and 31 percent with BMI>30.0), atherosclerosis (Prevalence: 30-40 percent), both hypertension (Prevalence: 18-25 percent) and hypotension (Prevalence: 10-20 percent), and diabetes mellitus (Prevalence: 10-17 percent=65 years). Most of these medical conditions require specific nutritional regimes for prevention and treatment. It is very likely that the optimization of nutrition in the general population and in specific population clusters with a particular risk associated with cerebrovascular disorders would help to reduce the prevalence and incidence of cases with cognitive deterioration compatible with either vascular or mixed dementia (7). Many studies reported during the past decade postulate the possibility that different diets or nutritional regimes may contribute to prevent diverse medical conditions and neurological disorders. Some examples include olive oil, flavonoids present in wine, tea, fruits and vegetables, vitamins, different types of antioxidants, polysaturated (PUFA) and monounsaturated fatty acids (MUFA), fish oil, marine lipoproteins, some trace elements and metals (iron, copper, zinc, selenium, silicon, lithium), and several herbal medicines (phytotherapy). PUFA are essential structural components of the CNS. PUFA-enriched diets lead to significant changes in expression of several genes in the brain (25). Deficiencies in essential long chain PUFAs (omega-3, omega-6) may result in cognitively impaired, suggesting that dietary lipids influence gene and protein activity levels, protein modifications, lipid raft formation, and protein aggregation involved in processes associated with higher activities of the CNS (26). It has been suggested that natural transthyretin inducers, such as omega-3-rich fish oil or pure preparations of docosahexaenoic acid (DHA), could be of benefit in the prevention of AD (25). Epigenetic factors also represent an important pathogenic component of complex disorders. Epigenetic-mediated changes in gene expression in response to dietary and other environmental exposures appear to be a molecular mechanism linking environmental factors with the genome with repercussion in cell function and health or disease throughout the life course (27). The evidence of a direct link between increase genome/epigenome damage and risk for adverse health outcomes in cancer and age-related disorders is becoming increasingly stronger. Genome and epigenome biomarkers are sensitive indicators of deficiency in micronutrients required as cofactors/components of DNA repair enzymes, for maintenance methylation of CpG islands and prevention of DNA oxidation and uracil incorporation into DNA (17). Despite this scattered information, the fact is that on a practical ground the nutrigenomics of dementia is still in its infancy; however, during the past few years novel classes of nutraceuticals have been introduced in the market, showing a genotype-related biopharmaceutical profile.

**Some genes associated with pharmacogenomic responses also influence nutrigenomics in dementia**

**Patients harbouring the APOE-4/4 genotype are the worst responders to both conventional treatments and specific nutrigenomic regimes**

**NUTRACEUTICALS IN LIPID METABOLISM DYSFUNCTION, CEREBROVASCULAR DISORDERS AND DEMENTIA**

Both vegetal and marine products are major sources of nutrients for humans. Different fish species exhibit diverse physiological responses to the environment that correspond to their phylogenetic grouping and evolutionary status, and constitute an optimal ecologic niche to investigate primitive molecules that underwent evolutionary changes with potential repercussion in health and disease in other evolved species. Recently, a series of novel nutraceutical products have been developed as marine derivatives by biotechnological procedures with potential application in lipid metabolism disorders, cancer, and dementia (5, 28, 29). Fish and fish oil consumption might encourage brain development and gene expression to brain maintenance during aging through nutrigenomic mechanisms (30). By using novel biotechnological procedures it is possible to obtain purified extracts of fish lipoproteins with atheroprotective and lipid-lowering effects or powerful immune system enhancers, as nutraceuticals with preventive and therapeutic applications (4, 5, 28, 29). E-SAR-94010 (LipoEsar) is a natural product extracted from the marine species S. pilchardus, by means of non-denaturalizing biotechnological procedures (28). The main chemical compounds of LipoEsar are PUFAs and lipoproteins (60-80 percent). In preclinical studies, LipoEsar has shown to be effective in (a) reducing blood cholesterol (CHO), triglyceride (TG), uric acid (UA), and glucose (Glu) levels, as well as liver alanine aminotransferase (ALT), and aspartate aminotransferase (AST) activity; (b) enhancing immunological function by regulating both lymphocyte and microglia activity; (c) inducing antioxidant effects mediated by superoxide dismutase activity; and (d) improving cognitive function (28, 29). Clinical studies have revealed that LipoEsar reduces the plasma levels of total cholesterol (T-CHO)(20-30 percent), Glu (5-10 percent), UA (10-15 percent), TG (30-50 percent), ALT and AST, after 1-3 months of treatment at a daily dose of 250-500 mg (t.i.d). The effect on T-CHO is the result of decreasing LDL-CHO levels and increasing HDL-CHO levels in parallel with an improvement in hepatic protection reflected by a reduction in ALT, AST, and GGT activity, as the result of reducing liver steatosis. The influence of different APOE genotypes on the therapeutic response of LipoEsar in patients with dementia and chronic dyslipidemia has been investigated (29). After one
month of treatment, LipoEsar (750 mg/day, p.o.) significantly reduced the serum levels of Glu, T-CHO (Figure 1), LDL-CHO, TG, urea, UA, ALT, AST, and GGT. These therapeutic outcomes were APOE genotype-related. The best responders were patients with APOE-3/3>APOE-3/4>APOE-4/4 (Figure 2)(29). The daily administration of 1000-1500 mg/day of E-SAR p.o. for 3 months also tended to reduce the average size of atherosclerotic plaques on the abdominal aortic wall by 10 percent. This effect is more significant in patients harboring the APOE-3/3 than in APOE-3/4 carriers in whom the size of the plaque is approximately 30-40 percent larger than in APOE-3/3 carriers (5). The biological properties of E-SAR-94010 represent a clear example of the potential benefits of nutraceutical intervention on lipid metabolism and cardio-cerebrovascular protection, also illustrating how the individual genetic profile affects the metabolic/therapeutic response to nutritional factors regulating lipid metabolism, as an extension of nutrigenomics (5).

FUTURE DIRECTIONS

The natural course of technical events to achieve efficient goals in pharmacogenomics and nutrigenomics include the following steps: (a) genetic testing of mutant genes and/or polymorphic variants potentially involved in nutritional process and food metabolism; (b) genomic screening, and understanding of transcriptomic, proteomic, and metabolomic networks associated with bi-directional food-genome interactions; (c) functional genomics studies and genotype-phenotype correlation analysis to assess specific parameters; and (d) development of nutraceuticals with a nutrigenomic profile for a personalized nutrition, addressing food safety and efficacy issues (4, 5). To achieve a mature discipline of nutrigenomics in CNS disorders and dementia it would be convenient to accelerate the following processes: (a) educate physicians and the public on the use of genetic/genomic screening in daily clinical practice; (b) standardize genetic testing for major categories of diseases; (c) validate nutrigenomic procedures according to food/nutraceutical category and pathology; (d) regulate ethical, social, and economic issues; and (e) incorporate nutrigenomic procedures to both nutraceuticals in development and foods of massive consumption in the market to optimize nutrition in the general population and in specific risk groups. The incorporation of nutrigenetics/ nutrigenomics and pharmacogenomic/pharmacogenetic protocols in AD may foster nutritional and therapeuthic optimization by helping to develop cost-effective drugs and novel nutraceutical products, improving efficacy and safety, reducing adverse events and non-compliance, and cutting down unnecessary costs for industry and society (3-12).

REFERENCES AND NOTES

34. C. Duval, M. Müller et al., Biochim Biophys Acta, 1771, pp. 961-971 (2007).
44. C. Duval, M. Müller et al., Biochim Biophys Acta, 1771, pp. 961-971 (2007).