Medical Hypotheses 118 (2018) 29–33

Contents lists available at ScienceDirect

Medical Hypotheses

journal homepage: www.elsevier.com/locate/mehy

Phenylalanine hydroxylase: A biomarker of disease susceptibility in Parkinson's disease and Amyotrophic lateral sclerosis

Glyn B. Steventon[a,*], Stephen C. Mitchell[b]

[a] ADMET Solutions Ltd, Ivar Gardens, Basingstoke, Hampshire RG24 8YD, UK
[b] Imperial College, Department of Computational and Systems Medicine, Faculty of Medicine, South Kensington, London SW7 2AZ, UK

ARTICLE INFO

Keywords:
S-oxidation polymorphism
Phenylalanine hydroxylase
Parkinson's disease
Amyotrophic lateral sclerosis
Biomarker

ABSTRACT

The S-oxidation of S-carboxymethyl-L-cysteine has been reported previously to be a biomarker of disease susceptibility in Parkinson's disease and Amyotrophic lateral sclerosis. In this investigation, the original observations have been confirmed with the incidence of the poor metaboliser phenotype (no urinary recovery of S-oxide metabolites) being found to be 3.9% within healthy control population. However, 38.3% of the Parkinson's disease subjects and 39.0% of the Amyotrophic lateral sclerosis group were phenotyped as poor metabolisers. The consequent odds risk ratio of developing Parkinson's disease was calculated to be 15.5 (95% CI 9.5–25.3) and for Amyotrophic lateral sclerosis was 15.2 (95% CI 8.8–26.5). Thus, the possible role of the enzyme responsible for the S-oxidation biotransformation reaction, phenylalanine hydroxylase, must be further investigated to elucidate the mechanism(s) of toxicity in susceptible individuals displaying these diseases. A dual role potentially explaining of the role of phenylalanine hydroxylase as a biomarker of disease susceptibility is presented together with the observation that metabolomics is a possible way forward in the identification of potential pro-toxins/toxins in those individuals phenotyped as poor metabolisers (Controls, Parkinson's disease and Amyotrophic lateral sclerosis subjects).

Introduction

Parkinson's disease

Parkinson's disease (PD), is the most common movement disorder and is characterized primarily by the loss of dopaminergic neurons in the substantia nigra pars compacta leading to a dopamine deficiency in the striatum. The loss of regulation of the basal ganglia neurones accounts for the motor symptoms which include bradykinesia, hypokinesia, rigidity, resting tremor and postural instability. In addition to these typical motor symptoms, various none motor features may also develop, such as autonomic dysfunction, sleep disturbances, depression and cognitive impairment. A pathological trait of sporadic PD is the presence of proteinaceous deposits within the neuronal perykarya called Lewy bodies and neuronal processes called Lewy neurites, which are mainly composed of α-synuclein, ubiquitin, neurofilaments and molecular chaperones [1,2].

Little is known about the aetiopathogenesis of PD. The most common sporadic form of PD seems to be a complex multifactorial disorder with variable contributions of environmental factors and genetic susceptibility. Aging is the most important risk factor, thus with increasing average life expectancy the incidence and prevalence of PD will rise considerably in the near future. The major stepforward in PD research was the discovery of genes which are responsible for the familial forms of the disease, α- synuclein, LRRK2 parkin, PINK1 and DJ-1 [3–11]. Both sporadic and the monogenetic forms share important clinical, pathological and biochemical features, such as the progressive demise of dopaminergic neurones in the substantia nigra. Thus, any insights into the function and dysfunction of PD-associated gene products will help to uncover the underlying mechanisms leading to neuronal cell death. Evidence in the literature now indicates that PD-associated genes directly or indirectly impinge on mitochondrial integrity, thereby providing a link to pathophysiological alterations observed in sporadic PD [3–11].

Amyotrophic lateral sclerosis

Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disease involving both the upper motor neurons and lower motor neurones. Average disease duration is about three years, but it can vary significantly. Death usually results from compromise of the respiratory muscles. The prevalence of individuals with ALS is roughly

* Corresponding author.
E-mail address: glynsteventon@btinternet.com (G.B. Steventon).

https://doi.org/10.1016/j.mehy.2018.06.018
Received 18 April 2018; Accepted 19 June 2018

Table 1
% Median urinary recovery of SCMC and its metabolites in a 0–8 h urine collection in Controls, PD and ALS subjects. The reported values are the median (minimum–maximum) recovery values of the parent drug and its metabolites in a 0–8 h timed urine collection (08:00–16:00 h) following the oral dosing of subjects with 750 mg of S-carboxymethyl-L-cysteine at 08:00 h.

Subjects recovered	n	% Total recovered	% Sulfides recovered	% S-oxides recovered	S-oxidation Index*
Controls	701	51.3 (17.3-98.2)	36.7 (9.3-93.9)	11.8 (0.0-45.4)	3.2 (0.5-299.00)
PD	175	47.9 (22.9-94.2)	44.5 (9.3-90.1)+	0.8 (0.0-45.4)+	75.5 (0.5-459.0)+
ALS	105	48.5 (28.8-90.8)	45.0 (18.9-90.1)+	0.8 (0.0-32.0)+	68.8 (1.3-553.0)+

%Total recovery is the % total amount of SCMC, sulfide and S-oxide metabolites recovered in the 0–8 h urine collection.

* S-oxidation Index = 100-% urinary S-oxides recovered/%urinary S-oxides recovered.

+ $P < 0.001$, Kruskal-Wallis One Way Analysis of Variance on Ranks for both PD and ALS subjects compared to Controls.

4–8:100,000 [12] and is similar to the number of newly diagnosed individuals each year.

Like PD little is known about the aetiology of sporadic ALS with reports of environmental causes of ALS including mercury, manganese and farming products (fertilizers, insecticides, herbicides) and dietary factors [13]. Genetics also plays a role in the disease pathology with an estimated 10% of individuals with ALS having at least one other affected family member. The enzyme superoxide dismutase (SOD) is particularly associated with ALS. SOD1 pathogenic variants account for 20% of all familial ALS and approximately 3% of sporadic ALS [14,15]. Thus, the aetiology of sporadic ALS is now thought to be multifactorial with a combination of mitochondrial dysfunction, oxidative stress, glutamate excitotoxicity and environmental triggers being proposed as biomarkers of disease susceptibility [16,17].

Xenobiotic metabolism and degenerative neurological disease

Since 1988 publications have been appearing in the literature indicating that sporadic PD and ALS are associated with abnormalities in the S-oxidation of S-carboxymethyl-L-cysteine (SCMC) [18–21]. No real advances were reported after 2003 and like the ring of power in Tolkein's Lord of the Rings triology "… some things that should not have been forgotten were lost. History became legend. Legend became myth." The story of the S-oxidation polymorphism and PD and ALS slowly faded with time. However, following the identification of the enzyme responsible for the sulfur oxygenation of SCMC in rat, mouse, HepG2 cells, humans and cDNA expressed phenylalanine hydroxylase proteins new life has been given to this story [22–32].

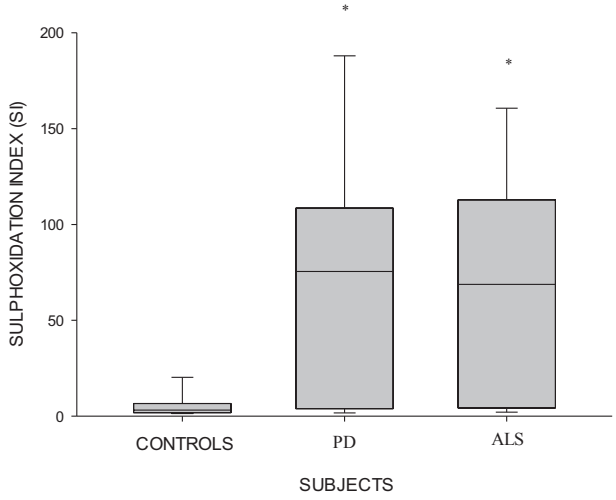

Fig. 1. Box plot of the S-oxidation Index in Controls, PD and ALS subjects. Subjects were phenotypes as previously reported [28,29]. S-oxidation Index = 100% – % urinary recovery of S-oxide metabolites/ % urinary recovery of S-oxide metabolites. Box plots show the median, the 10th-90th percentiles and the errors of the distributions *P < 0.001 Kruskal-Wallis One way analysis of variance on ranks.

Hypothesis

It is proposed that the enzyme known as phenylalanine hydroxylase (phenylalanine monooxygenase, PAH, EC. 1.16.14.1) whose classical function is the conversion of phenylalanine to tyrosine, has other metabolic roles within the body. Although phenylalanine is the preferred substrate the enzyme has been shown to oxidise several other compounds including the drug, S-carboxymethyl-L-cysteine. Whilst most allelic variants of PAH adequately metabolise phenylalanine, thereby avoiding any clinical consequence, they are unable to effectively metabolise these secondary substrates. Poor sulphoxidation of S-carboxymethyl-L-cysteine has been reported as a risk factor for developing PD and ALS, the drug being a presumed metabolic 'probe' for PAH allelic variants. It is proposed that these allelic variants of PAH, themselves capable of maintaining sufficient phenylalanine metabolism, are unable to metabolise other as yet unknown 'toxic substances', and it is the accumulation of subsequent 'toxic insults' that assist in the development of these neurological problems.

The S-oxidation polymorphism

A re-evaluation of the S-oxidation polymorphism in controls, PD and ALS subjects has been undertaken to investigate the association of PAH with these diseases. The collated data now re-evaluated has been published previously in various forms [18–21,32,33] but never has the

Table 2
S-oxidation phenotypes in Controls, PD and ALS subjects. The S-oxidation phenotype was determined for each subject using the antimode of < 1.6% urinary recovery of S-oxide metabolites. If a subject is reported has having < 1.6% urinary recovery of S-oxide metabolites in a 08:00–16:00 h urine collection, the recovered S-oxide metabolites are below the level of quantification.

Subjects	n	EM phenotype (%)	PM phenotype (%)	OR 95% CI	Z	RR	Z	NNH 95% CI
Controls	701	674 (96.1)	27 (3.9)					
PD	175	108 (61.7)*	67 (**38.3**)*	**15.5*** 9.5–25.3	10.9	**5.2***	14.8	1.7 1.5–2.0
ALS	105	64 (61.0)*	41 (**39.0**)*	**15.2***	9.6	**6.8***	12.2	2.0 1.7–2.3

EM = % Urinary recovery of S-oxides ≥1.6% or SI ≤ 80, PM = % Urinary recovery of S-oxides < 1.6% or SI > 80.
OR = Odds Ratio, RR = Relative Risk, NNH = number need to harm.
* $P < 0.0001$, Chi squared test with Yates' correction.