Research Article

STUDY OF LATENT EARLY-LIFE ASSOCIATED REGULATIONS AND AUTOMATED IDENTIFICATION OF PARKINSON'S DISEASE

Nivedita Chaudhary, Yogender Aggarwal and [*]Rakesh Kumar Sinha
*Center for Biomedical Instrumentation, Department of Electrical and Electronics Engineering
Birla Institute of Technology, Mesra, Ranchi, Jharkhand (India)-835215
*Author for Correspondence

ABSTRACT

In the present study, one model that has already been proposed for understanding the etiology of Alzheimer's disease, *i.e.*, 'Latent Early-Life Associated Regulation' (LEARn) model is studied for the Parkinson's disease (PD). LEARn model postulates latent expressions of specific genes triggered at the developmental stage. According to this model, environmental agents (e.g. heavy metals ions, pesticides, temperature and pH) disturb gene (primarily PARK 1) regulation in a long-term fashion, beginning at early developmental stages. Pathologically, α-synuclein is identified as the main culprit; oxidative stress and mitochondrial dysfunction play important role in onset of disease followed by degeneration of dopamine-producing brain cells. The clinical symptoms of PD has been studied with the help of an expert clinician and identified as it is expressed under four major classes of clinical symptoms: (i) Communication problems, (ii) Personality changes, (iii) Loss of voluntary control and (iv) Health problems. All these symptoms appear very late and it get worsen over time. In this work, an artificial neural network (ANN) has been used to identify the PD conditions with the help of clinical symptoms with an accuracy of 98%. The used symptomatic inputs to the ANN have provided very effective identification rate for PD conditions. Based on this study, it can be suggested that the outputs of biochemical network models can be combined with smart computational tools to get effective diagnostic system for neurodegenerative disorders.

Key Words: *Artificial Neural Network, Environmental Factors, Learn Model, Parkinson's Disease*

INTRODUCTION

Neurodegenerative diseases represent a large group of neurological disorders with heterogeneous clinical and pathological expression affecting specific subsets of neurons in definite functional anatomic systems: they arise for unknown reasons and progress in a relentless manner (Przedborski *et al.*, 2003). Parkinson's disease (PD) is the second most common neurodegenerative disease among elderly people (Takahashi & Imai, 2003). It is a chronic, progressive neurodegenerative disorder that affects at least 1% of the population by age 70 (Savitt *et al.*, 2006), regarded as the loss of dopamine-containing neurons in the substantia nigra pars compacta (SNPC) (Brochard *et al.*, 2009) and clinically described by the symptoms of bradykinesia, resting tremor, rigidity, postural instability, autonomic, cognitive, and psychiatric disturbances (Moore *et al.*, 2005). Basically two main theories are considered for pathogenesis of PD (Dauer and Przedborski, 2003). One hypothesizes that misfolding and aggregation of proteins are influential in the loss of SNPC dopaminergic neurons, while the other is based on the idea that irregular mitochondrial structure and function contributes to increased oxidative stress and toxic dopaminergic species. PD pathogenesis also includes abnormalities in cellular protein transport, interaction between proteins, mitochondrial dysfunction, oxidative stress as well as the genetic mutation. These processes are considered to form a complex cascade of unified events that lead to neuronal death by the manner of apoptosis.

In humans, appearances of PD usually start in latter half of life. It is characterized by progressive loss of muscle control, stiffness, slowness, and impaired balance (Fahn, 2003). All these symptoms appear very late and it get worsen over time and as symptoms appear, 60-70% of the striatal dopaminergic terminals have been already lost (Drechsel and Patel, 2008). It is still not clear when this disease actually starts and

Research Article

how long it will take to be visible symptomatically. Thus, development and progression of PD with the help of biochemical models is difficult. Considering this limitation, the established model for understanding the etiology of Alzheimers disease, the 'Latent Early-Life Associated Regulation' (LEARn) model (Lahiri *et al.*, 2007; 2008; Lahiri and Maloney, 2010) has been considered for the explanation of PD. This model suggests a latent expression of specific genes triggered at the developmental stage. According to this model, environmental agents disturb gene regulation in a long-term fashion, beginning at early developmental stages.

It is revealed that multiple symptoms arise progressively that may be indicative of PD. Thus, its automated differentiation with normal subjects is important as the manual screening is a tedious and error frame task. The proposed artificial neural network (ANN) model is based on a set of multilayered interconnected equations, which uses non-linear statistical analysis to reveal previously unrecognized relations between given input variables (assigned according to the severity of expressed symptoms) along with an output variable. Several studies have shown that ANN is accurate and reliable in diagnosis and outcome prediction in diverse clinical setting (Sinha, 2008). ANNs also appear to be promising for classification of neurodegenerative disorders (Litvan *et al.*, 1996; Ene, 2008). In near past, its application is also reported in identification of the metabolic marker for the detection of PD from plasma and to validate the variations by spotting out through gene responsibility (Ahmed *et al.*, 2009).

In this paper, the biochemical pathway of PD has been studied by using the LEARn model. The proposed LEARn model for PD is based on environmentally induced hypomethylation or oxidative damage as the physical mechanism that perturbs gene expression and considers the effects of environmental factors (heavy metals ions, pesticides, temperature and pH) on both oxidative stress and genetic mutation (PARK 1) associated with this disease. Further, apart from LEARn model, the present work is aimed to develop an ANN model to predict the appearance of PD by use of the symptoms routinely available to the clinicians.

ETIOLOGY OF PARKINSON'S DISEASE

PD arises from genetic and possibly neurotoxic causes that produce massive cell death of the neuromelanin-containing dopaminergic neurons of the substantia nigra (Sulzer, 2011). Substantia nigra degeneration is one of the major features of PD because symptoms only get visible when most of the cells are dead in this region. There was a decline in axonal transport motor proteins in sporadic PD that preceded other well known nigral cell-related pathology such as phenotypic down regulation of dopamine (Chu *et al.*, 2012). Reduced axonal transport contributes to the degeneration of neuronal processes in PD. Mitochondria supply the ATP needed to support axonal transport and contribute to many other cellular functions essential for the survival of neuronal cells. Oxidative stress causes mitochondrial dysfunction and can impair proteasomal function. Impairment of proteasomal function induces oxidative and nitrative stress and inhibits mitochondrial function (Jenner, 2003) that impairs protein clearance as ATP production is required for normal proteolytic activity. Along with all pathological pathways, age cannot be denied as a contributing factor for PD and loss of dopaminergic neurons seen in PD can be distinguished from normal changes associated with aging. In those with PD, neuronal cell death occurs mainly in ventrolateral and caudal portions of the SNPC. In normal aging, these changes are seen in the dorsomedial aspect of the SNPC (Dauer & Przedborski, 2003).

Peptide Aggregation

Protein aggregation is a common feature of many neurodegenerative diseases, and it is assumed that the aggregation process plays a central role in pathogenesis. In this process, the monomer of a soluble protein interacts with other monomers of the same protein to form dimers, oligomers, and polymers. As the size of the aggregates increases, they may precipitate as insoluble amyloid fibrils (Irvine *et al.*, 2008). One of the pathological hallmarks of PD, Lewy bodies (LB) are spherically-shaped eosinophillic cytoplasmic protein aggregates found in all affected brain regions that are made up of several proteins, including α-synuclein, parkin, ubiquitin, and neurofilaments. In PD, LBs contain α-synuclein that has been altered by

Research Article

oxidative activity. The α-synuclein was identified as the major component of amyloid fibrils found in LB body and Lewy neurites, the characteristic proteinaceous deposits that are the diagnostic hallmarks of PD. In vitro studies have shown to this form of the protein demonstrates a greater tendency to aggregate than unmodified α-synuclein, which supports the theory that oxidative stress plays a key role in PD pathogenesis. It is an intrinsically unfolded protein that can adopt a partially helical structure when it interacts with different lipid membranes. The amino-terminal sequence of α-synuclein is almost entirely composed of variants of an imperfect 11 amino acid repeat (XKTKEGVXXXX) (Bisaglia *et al.*, 2009).

Genetic Causes

Most cases of PD arise sporadically, but newer etiological research is discovering a smaller number of cases caused by genetic mutations (de Lau & Breteler, 2006). PD was thought, until recently, to have little or no genetic component. This notion has changed with the identification of three genes, and the mapping of five others, that are linked to rare familial forms of PD (FPD). Recent evidences suggest the involvement of single gene mutations including α-synuclein (PARK 1), Parkin (PARK 2), and ubiquitin-C-hydrolase-L1 (PARK 5); LRRK 2, DJ-1, PARK 3, PINK 1, and HTRA 2 are the underlying mechanism of FPD pathogenesis. There is evidence that mutation of the gene and over expression of α-synuclein is associated with decreased mitochondrial function, abnormal mitochondria, damaged mitochondrial DNA, increased cytochrome c release, and increased free radical production (Henchcliffe & Beal, 2008). PINK 1 gene encodes a putative serine/threonine protein kinase with a mitochondrial import sequence located at its N-terminus (Yao *et al.*, 2011). PINK 1 interacts with Parkin to regulate mitochondrial dynamics, including fission/fusion. PINK 1 initiates mitophagy via phosphorylation of Parkin. Its stabilization on the outer mitochondrial membrane is necessary for Parkin ligase, which is normally localized to the cytosol, to translocate to damaged mitochondria. PINK 1 mutation that is related to PD pathology impairs mitochondrial autophagy by compromising initial Parkin translocation to mitochondria. In normal circumstances, PINK 1 recruits Parkin following a loss of mitochondrial membrane potential (Geisler *et al.*, 2010). DJ-1 is normally localized in the cytosol, but its expression is noticed in the mitochondria under conditions of oxidative stress and has been found to play a significant part in the defensive cellular response to oxidative stress. It is also suggested that this could work upstream of the PINK 1/Parkin activation or it could work within an independent, but parallel, pathway in conjunction with PINK 1/Parkin. DJ-1 deficient cells also have lower mitochondrial fusion, an increased tendency to fragment, and impaired mitochondrial dynamics leading to an increase in autophagy. This also suggests that DJ-1 function in normal cells could regulate autophagy or attenuate the downstream negative effects of reactive oxygen species (Thomas *et al.*, 2011). LRRK 2 is associated with idiopathic PD. It is typically late-onset PD LRRK 2, also called leucine-rich repeat serine/threonine-protein kinase 2, is localized in the cytoplasm. Mutations of LRRK 2 cause PARK 8 (FPD, type 8) and augment MAPK activity. It causes an increase in phosyphorlation intra and inter-cellularly (Farrer & Ross, 2006). The pathology of LRRK 2 includes deposits of aggregated protein (Smith *et al.*, 2005). It is rare to find mutations of the HTRA 2 gene in individuals with PD. Mutations of the gene result in increased mitochondrial volume, electrochemical changes in the mitochondrial membrane, and increased risk of cell death related to the activity of staurosporine, an ATP kinase inhibitor. It is also thought that mutations to the PINK 1 gene negatively influence HTRA 2 function (Henchcliffe & Beal, 2008).

Environmental Risk Factors

Several studies have demonstrated that environmental factors implicated in PD, including heavy metals and pesticides, accelerate *in vitro* fibril formation. Epidemiological studies have demonstrated that the following environmental factors are positively correlated with the risk of developing PD: pesticide use for example rotenone and paraquat (Tansey *et al.*, 2007), contact with certain industrial chemicals (trichloroethylene, manganese, carbon disulfide, carbon monoxide, cyanide, methanol), exposure to wood preservatives, drinking well water (Priyadarshi *et al.*, 2001), and exposure to MPTP (Ballard *et al.*, 1985). There are thoughts that some of these environmental factors contribute to the mitochondrial dysfunction seen in primary PD (Dick, 2006). One of the strongest relationships reported in the literature is the

Research Article

negative correlation between cigarette smoking (Baron, 1986) and caffeine intake and risk of PD development (Hernán *et al.*, 2002; Checkoway *et al.*, 2002).

LEARn Model and Its Implications

A 'Latent Early-life Associated Regulation' (LEARn) model, postulates a latent expression of specific genes triggered at the developmental stage. It is already proposed that the majority of Alzheimer's disease cases would, instead, follow an etiology based on 'LEARn' (Lahiri *et al.*, 2007; 2008; Lahiri and Maloney, 2010). According to that, LEARn posits that early-life influences, such as exposure to metals, nutritional variation, variation in maternal care, and other stressors modify potential expression levels of disorder-associated genes in a latent fashion. Such latent changes are maintained by epigenetic markers in the promoter sequences of such genes. Based on this hypothesis, in the present work, we integrated both the reasons of PD; the neuropathological features (e.g., α-synuclein and LB) as well as the oxidative stress effected by environmental factors such as Pesticides, metal ions exposure (Uversky *et al.*, 2001; Willis *et al.*, 2010), pH and temperature (Mcnulty *et al.*, 2006). Environmental agents perturb gene regulation in a long-term fashion, beginning at early developmental stages, but these perturbations do not have pathological results until significantly later in life.
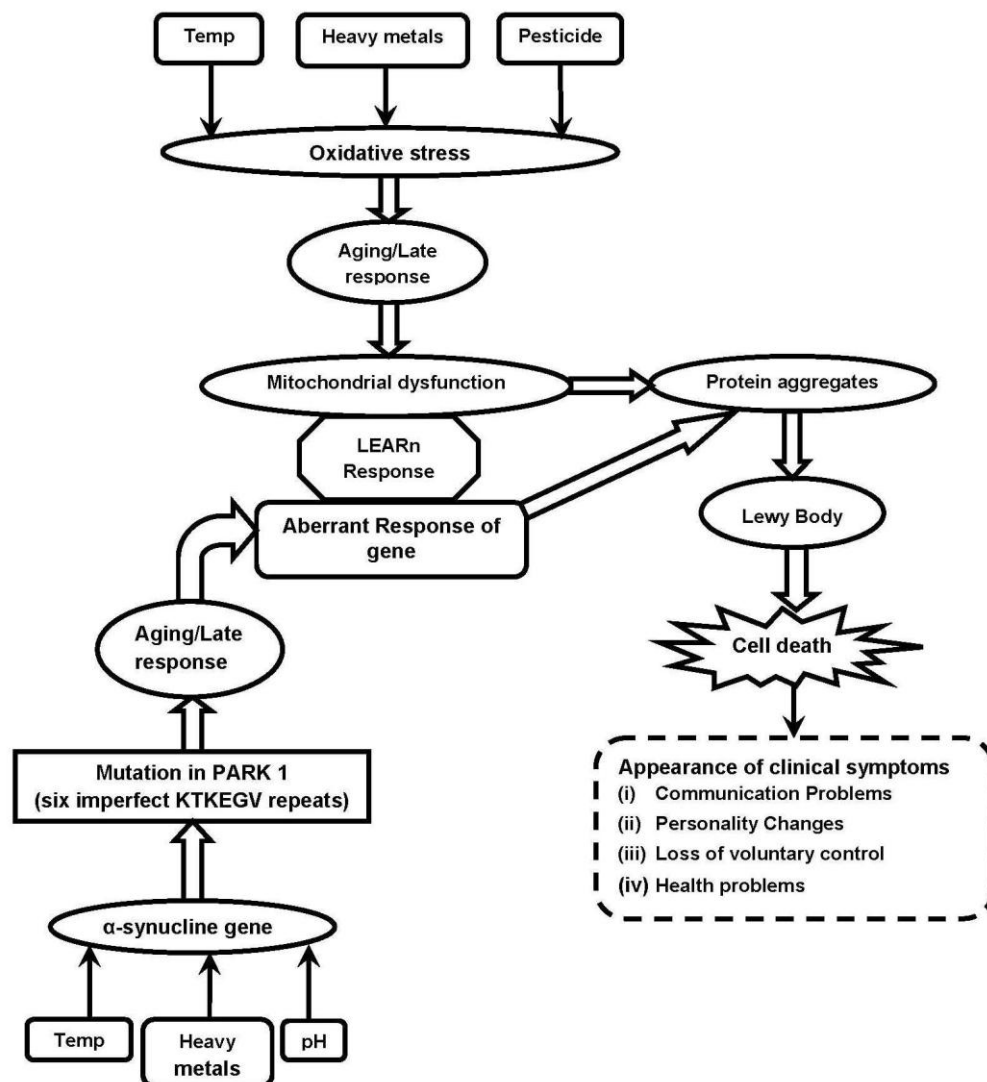

Figure 1: Block diagram of Latent Early-life Associated Regulation (LEARn) model

Research Article

Like all other Neurodegenerative diseases PD apparent very late in adult life, it is still not clearly understood that when the disease actually starts and how long the biochemical events and neuropathological processes take to develop the disease. So the major unresolved question is related to the time and aging. There are multiple pathways contributing to PD, and there are several indications that environmental factors affecting these pathways in various manners (Uversky *et al.*, 2001; Meigal & Lupandin, 2005; Coon *et al.*, 2006). Therefore, to explain the etiology of PD with the help of LEARn model, we considered effect of various environmental factors associated with PD. As the LEARn model is based on environmentally induced hypomethylation or oxidative damage that effect the gene expression (Bolin *et al.*, 2006), these perturbations are latent. They are not immediately apparent in the same manner found in conventional toxic responses.

Peptide aggregation is major cause of PD. In the present model, both the causes are considered genetic mutation and result of oxidative stress due to environmental insult such as temperature, heavy metals, pH and pesticides. Genetic mutation initiated by environmental insult causes six imperfect repeats in PARK 1 gene this is immediate effect on gene but the symptoms only get visible after a long time or aging when due to this mutation peptide aggregation starts here that can be noticed in the LEARn response. Simultaneously, it is also shown that oxidative stress caused by environmental factors leads to mitochondrial dysfunction as an immediate response but after LEARn response it also results in peptide aggregation, which ultimately leads to cell death and neurodegeneration. The LEARn model proposes that the initial triggering mechanism activates early in life, but the development of disease occurs very late. Figure-1 explains the pathways of LEARn response.

CLINICAL SYMPTOMS BASED ANN IDENTIFIER FOR PARKINSON'S DISEASE

As discussed in LEARn response (figure-1), with peptide aggregation that leads to neuronal cell death, the clinical symptoms of PD appear very late, usually in the latter half of the life. The clinical symptoms of PD has been studied with the help of an expert clinician and identified as it is expressed under four major classes: (i) Communication problems, (ii) Personality changes, (iii) Loss of voluntary control and (iv) Health problems. The characteristics of the PD under each of the mentioned major classes are further focused on the specific symptoms and explained in table-1. Appearance of majority of these clinical symptoms, if not all, is used as confirmatory parameters in clinical domain.

Based on clinical symptoms, the present paper has used three layered feed-forward back propagation ANN to differentiate PD from the normal condition. The ANN network was coded using C++ programming language (Rao and Rao, 1996). The backpropagation ANN has advantage of available effective training and better-understanding of system behavior. Neurons or nodes in each layer are interconnected in a feed-forward fashion. The connections between different layers of nodes have associated weights, which act upon outputs of the first layer of nodes before they are processed next. In the present network, the nodes of hidden and output layers have a sigmoid transfer function (Aggarwal *et al.*, 2007). The weights were adjusted as follows:

$$W_{ij}(t+1) = W_{ij}(t) + \eta \delta_j X_i + \alpha(W_{ij}(t) - W_{ij}(t-1)) \tag{1}$$

Where $W_{ij}(t)$ is the connection weight from a node i in one layer at time t, X_i is either an input or the output of the hidden node i, δ_j is an error term for node j, η is a learning rate factor and α is a momentum factor $(0 < \alpha < 1)$. If j is an output node, then

$$\delta_j = y_j(1-y_j)(d_j - y_j) \tag{2}$$

Where, d_j is the desired output of node j and y_j is the actual output. If node j is hidden node, however, the computation of error terms becomes

$$\delta_j = X_j(1-X_j)\sum_k \delta_k W_{jk} \tag{3}$$

Research Article

Where k is summed over all nodes in the layer above node j. Hidden node thresholds were adjusted in a similar way by assuming they are connection weights on links from auxiliary constant valued inputs.

In ANN, digital values of the PD symptoms (table-1) were used as the input vector to the ANN. The inputs have been manually prepared in digital form in the scale between 0 and 1. The severity of any symptom has an assigned value > 0.5 and for the normal conditions, it is assigned < 0.5. Similarly, the PD condition was predicted with the firing intensity of the output node. The output value > 0.5 of output node confirms the PD and the increasing intensity of firing of output node provides the severity of PD condition in digital form. In the present study, 160 data sets (80 normal and 80 PD) have been prepared with the help of expert clinician to test the ANN. The 13:20:1 architecture of ANN was obtained with the optimization both for training and testing purpose. During training, a training file 'TRAINING.DAT' with the digital values of all thirteen symptoms along with an output was presented to the input. For training of network 60 datasets (30 each for normal and PD) were randomly arranged. Remaining 100 data sets (50 each for normal and PD) were used for testing purposes. The error tolerance, number of iterations and learning rate parameter were assigned as 0.01, one million and 0.1, respectively. Once the simulator reaches the error tolerance specified or achieved the maximum number of iterations, assigned for training, the simulator save the state of network by saving all its weights in 'WEIGHT.DAT' file. The output values have been compared to the desired outputs and if the outputs have found same as desired outputs, no further training is required. Otherwise weights have to adapt further by training with least mean square rule. Obtaining the error in minimum possible value, results in absolute correction. During testing, the testing file 'TEST.DAT' with digital values of clinical symptoms is presented to the input nodes of ANN, which read the input and finds the output of the network.

Table 1: Clinical variable of Parkinson's disease used to build the predictive model.

Major Clinical Variables	Clinical Stages	Primary Symptoms	Secondary Symptoms
Communication Problems	Conversation Comprehension Slow response		Soft whispery voice Small cramped handwriting Slow response to questions
Personality Changes	Depression Mental disorders		Depression Anxiety, Isolation
Loss of voluntary control	Bradykinesia	Slowness in voluntary movements such as standing up, walking and sitting down, freezing episodes	
	Tremors	Often occur in the hands, fingers, forearms, foot, mouth or chin	
	Rigidity	Stiff muscles, muscles pain during movement	
	Poor balance	Unsteady balance, which oftentimes leads to fall, loss of reflexes that help posture	
Health problems	Loss of bowel and bladder control		Constipation, loss of bowel and bladder control
	Throat, mouth and esophagus complications		Difficulties in swallowing, choking, coughing or drooling, excessive salivation
	Dermatological effects		Excessive sweating, scaling, dry skin on the face or scalp
	Dependence		At advanced stage needs complete assistance

Research Article

Before applying the ANN for the identification of PD, the network was optimized to get optimum performance. Different learning rates were investigated in the range of 0.01-0.5. The best performance was when the learning rate was chosen as 0.1 with which the accuracy of 100% was obtained for training set. The effects of number of hidden nodes in the range of 5-50 with a fixed number of iterations (1 million) are tested. 20 hidden nodes resulted in the best performance compared with other combinations of hidden nodes. With the 13:20:1 structure and optimized learning rate value (0.1), the ANN classified 50 normal and 50 diseased test sets with the accuracy of 98%.

DISCUSSION
Like other neurologic diseases, PD is related to aging, may be determined by chronic exposure of adverse environmental factors early during the life span or even during pregnancy (Logroscino, 2005). Earlier the etiologic model for PD has explored mainly risk factors associated with adult life or in many cases after the age of 65 (Kuh & Ben-Shlomo, 1997). The data for PD and AD suggest that a number of environmental insults occurring early in life may lead or contribute to these diseases (Landrigan *et al.*, 2005). PD also fits in two-hit disorder, similarly to those found in currently accepted models of cancer etiology (Knudson, 1971) and Alzheimer's disease (Lahiri and Maloney, 2010).
LEARn mechanism reveals the interaction between oxidative stress, gene and environmental factors in early developmental etiology for PD. The promoter regions of gene PARK 1 provide the substrate for LEARn activity. Environmental factors such as exposure to temperature, heavy metal and pH result in six imperfect repeats in PARK 1 gene and also the effect of environmental factors (temperature, heavy metals and pesticides) on oxidative stress that alter the function of mitochondria, which ultimately results in peptide aggregation and cell death, both the pathways contribute to neurodegeneration. In LEARn systems, alterations are latent and effects are primarily notable later in life. In both immediate and LEARn systems, epigenetic changes in gene regulation result in disorders. However, the LEARn model has its limitations as it does not provide any predictive output to analyze the severity of the disorder. PD is age-associated progressive neurodegenerative disease that gradually affects human brain. It is already known that the disease starts early in the life but the symptoms appear very late. LEARn model is only to explain that what the factors are affecting in our early life which later results in to the disease and to explain that there is the latent period between immediate effect and symptoms to be get visible. This model does not explain that when during the life course that a given exposure has its greatest effect and how exposures may accumulate over the life span. More elaborative and ultimate knowledge of risk factors involved will be needed to implement intervention and preventative strategies early in life to reduce or prevent any adverse late-life outcomes. However, the LEARn model will help to efficiently detect these changes and early-life remediation methods to eliminate PD before it occurs. We extended the Lahiri's LEARn model (Lahiri *et al.*, 2007; 2008; Lahiri & Maloney, 2010) by adding symptomatic classification of Parkinson's after onset of disease with the help of ANN.
In this study, we used ANN based technique is an attempt to envisage the severity of PD. For any predictive technique to be useful in making a diagnostic decision, an important feature is that only data that are readily available to the clinician at the time of diagnosis are used. Our model used only data derived from clinical history and physical examination. We accentuate that our model is not meant to replace or substitute for an experienced clinician; on the contrary, we suggests that the model should be viewed as a decision aid for the busy clinicians. The result of this study are promising, suggesting that a system based on this approach will be accurate and user friendly, while simple enough to be implemented at low cost.
The review of literature revealed that the ANN is being applied in broad areas of prediction and classification. In the field of health and medicine it is used by many researchers for diagnosis and prediction of diseases. Neural networks are powerful computational methods that have resulted from work in the area of neurophysiology and are being applied to an increasing range of medical problems (Paliwal & Kumar, 2009). In the field of neurodegenerative diseases ANN is applied for classification of

Research Article

cognitively normal, demented, AD and vascular dementia from single photon emission with computed tomography image data from brain (deFigueiredo *et al.*, 1995), to distinguish AD patients from controls in the nun study (Grossi *et al.*, 2007), also for the classification of essential tremor, PD tremor, and physiological tremor (Ai *et al.*, 2008). In 1996, Litvan and co-workers applied supervised and unsupervised ANN for differentiating the subtypes of progressive supranuclear palsy (PSP), and in differentiating PSP from postencephalitic Parkinsonism (PEP) and corticobasal degeneration, or Pick's disease from corticobasal degeneration. The supervised learning ANN achieved excellent accuracy in classifying PSP but had difficulty in classifying the other disorders. There have been no reports on the application of ANN methods for symptomatic diagnostic classifications of PD with the help of symptoms regularly available to the clinicians. Further, there are no clinically useful predictive techniques available to aid the physician in the diagnosis of patients. Therefore, it can be suggested that the prediction of severity with the help of ANN following the understanding of LEARn response of PD may provide a useful tool in clinical domain.

Conclusion
PD is a progressive neurodegenerative disorder manifested by a broad spectrum of motor and non-motor features. Occurrence of PD has been reported to be associated with environmental factors by many researchers. LEARn model along with the ANN classifier is not only useful for diagnosing the disease in affected persons, it will be useful for identifying family members or populations at risk, thus providing an opportunity to initiate neuroprotective therapy at an asymptomatic stage.

ACKNOWLEDGEMENT
Conflict of Interest
Authors of the manuscript certify that there is no ethical issue or conflict of interest involved in this work. Further, they have no financial interests related to the material in the manuscript.

REFERENCES
Aggarwal Y, Karan BM, Das BN, Aggarwal T and Sinha RK (2007). Backpropagation ANN-based prediction of exertional heat illness. *Journal of Medical Systems* **31**(6) 547-550.
Ahmed SS, Santosh W, Kumar S and Christlet HT (2009). Metabolic profiling of Parkinson's disease: evidence of biomarker from gene expression analysis and rapid neural network detection. *Journal of Biomedical Science* **16** 63.
Ai L, Wang J and Wang X (2008). Multi-features fusion diagnosis of tremor based on artificial neural network and D–S evidence theory. *Signal Processing* **88**(12) 2927-2935.
Ballard PA, Tetrud JW and Langston JW (1985). Permanent human parkinsonism due to 1-methy l-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP): seven cases. *Neurology* **35**(7) 949-956.
Baron JA (1986). Cigarette smoking and Parkinson's disease. *Neurology* **36**(11) 1490-1496.
Bisaglia M, Mammi S and Bubacco L (2009). Structural insights on physiological functions and pathological effects of alpha-synuclein. *FASEB Journal* **23**(2) 329-340.
Bolin CM, Basha R, Cox D, Zawia NH, Maloney B, Lahiri DK and Cardozo-Pelaez F (2006). Exposure to lead and the developmental origin of oxidative DNA damage in the aging brain. *FASEB Journal* **20**(6) 788-790.
Brochard V, Combadière B, Prigent A, Laouar Y, Perrin A, Beray-Berthat V, Bonduelle O, Alvarez-Fischer D, Callebert J, Launay J, Duyckaerts C, Flavell RA, Hirsch EC and Hunot S (2009). Infiltration of CD4+ lymphocytes into the brain contributes to neurodegeneration in a mouse model of Parkinson disease. *Journal of Clinical Investigation* **119**(1) 182-192.
Checkoway H, Powers K, Smith-Weller T, Franklin GM, Longstreth WT Jr and Swanson PD (2002). Parkinson's disease risks associated with cigarette smoking, alcohol consumption, and caffeine

Research Article

intake. *American Journal of Epidemiology* **155**(8) 732-738.

Chu Y, Morfini GA, Langhamer LB, He Y, Brady ST and Kordower JH (2012). Alterations in axonal transport motor proteins in sporadic and experimental Parkinson's disease. *Brain* **135**(7) 2058-2073.

Coon S, Stark A, Peterson E, Gloi A, Kortsha G, Pounds J, Chettle D and Gorell J (2006). Whole-body lifetime occupational lead exposure and risk of Parkinson's disease. *Environmental Health Perspectives* **114**(12) 1872-1876.

Dauer W and Przedborski S (2003). Parkinson's disease: mechanisms and models. *Neuron* **39**(6) 889-909.

De Lau LM and Breteler MM (2006). Epidemiology of Parkinson's disease. *Lancet Neurology* **5**(6) 525-535.

De Figueiredo RJ, Shankle WR, Maccato A, Dick MB, Mundkur P, Mena I and Cotman CW (1995). Neural-network-based classification of cognitively normal, demented, Alzheimer disease and vascular dementia from single photon emission with computed tomography image data from brain. *Proceedings of the National Academy of Science of the United States of America* **92**(12) 5530-5534.

Dick FD (2006). Parkinson's disease and pesticide exposures. *British Medical Bulletin* **79-80**(1) 219-231.

Drechsel DA and Patel M (2008). Role of reactive oxygen species in the neurotoxicity of environmental agents implicated in Parkinson's disease. *Free Radical Biology and Medicine* **44**(11) 1873-1886.

Ene M (2008). Neural network-based approach to discriminate healthy people from those with Parkinson's disease. *Annals of the University of Craiova, Mathematics and Computer Science Series* **35** 112-116.

Fahn S (2003). Description of Parkinson's disease as a clinical syndrome. *Annals of the New York Academy of Sciences* **991**(1) 1-14.

Farrer M and Ross O (2006). LRRK2-related parkinson disease, LRRK2 associated parkison disease, PARK8. Gene Reviews, NCBI Bookshelf. A service of the National Library of Medicine, National Institutes of Health. Available: http://www.ncbi.nlm.nih.gov/books/NBK1208/.

Geisler S, Holmstrom KM, Treis A, Skujat D, Weber SS, Fiesel FC, Kahle PJ and Springer W (2010). The PINK1/Parkin-mediated mitophagy is compromised by PD-associated mutations. *Autophagy* **6**(7) 871-878.

Grossi E, Buscema MP, Snowdon D and Antuono P (2007). Neuropathological findings processed by artificial neural networks (ANNs) can perfectly distinguish Alzheimer's patients from controls in the Nun Study. *BMC Neurology* **7** 15.

Henchcliffe C and Beal MF (2008). Mitochondrial biology and oxidative stress in Parkinson disease pathogenesis. *Nature Clinical Practice Neurology* **4**(11) 600-609.

Hernan MA, Takkouche B, Caamano-Isorna F and Gestal-Otero JJ (2002). A meta-analysis of coffee drinking, cigarette smoking, and the risk of Parkinson's disease. *Annals of Neurology* **52**(3) 276-284.

Irvine GB, El-Agnaf OM, Shankar GM and Walsh DM (2008). Protein aggregation in the brain: the molecular basis for Alzheimer's and Parkinson's diseases. *Molecular Medicine* **14**(7-8) 451-464.

Jenner P (2003). Oxidative stress in Parkinson's disease. *Annals of Neurology* **53 Suppl 3** S26-S38.

Knudson AG Jr (1971). Mutation and cancer: statistical study of retinoblastoma. *Proceedings of the National Academy of Science of the United States of America* **68**(4) 820-823.

Kuh D and Ben-Shlomo Y (1997). A Life Course Approach to Chronic Disease Epidemiology. *Oxford University Press,* New York.

Lahiri DK, Maloney B, Basha MR, Ge YW and Zawia NH (2007). How and when environmental agents and dietary factors affect the course of Alzheimer's disease: the "LEARn" model (latent early-life associated regulation) may explain the triggering of AD. *Current Alzheimer Research* **4**(2) 219-228.

Research Article

Lahiri DK, Zawia NH, Greig NH, Sambamurti K and Maloney B (2008). Early-life events may trigger biochemical pathways for Alzheimer's disease: the "LEARn" model. *Biogerontology* **9**(6) 375-379.

Lahiri DK and Maloney B (2010). The "LEARn" (Latent Early-life Associated Regulation) model integrates environmental risk factors and the developmental basis of Alzheimer's disease, and proposes remedial steps. *Experimental Gerontology* **45**(4) 291-296.

Landrigan PJ, Sonawane B, Butler RN, Trasande L, Callan R and Droller D (2005). Early environmental origins of neurodegenerative disease in later life. *Environmental Health Perspectives* **113** (9) 1230-1233.

Litvan I, Deleo JM, Hauw JJ, Daniel SE, Jellinger K, McKee A, Dickson D, Horoupian DS, Lantos PL and Tabaton M (1996). What can artificial neural networks teach us about neurodegenerative disorders with extrapyramidal features? *Brain* **119**(Pt 3) 831-839.

Logroscino G (2005). The role of early life environmental risk factors in Parkinson disease: what is the evidence? *Environmental Health Perspectives* **113**(9) 1234-1238.

McNulty BC, Tripathy A, Young GB, Charlton LM, Orans J and Pielak GJ (2006). Temperature-induced reversible conformational change in the first 100 residues of alpha-synuclein. *Protein Science* **15** (3) 602-608.

Meigal A and Lupandin Y (2005). Thermoregulation-dependent component in pathophysiology of motor disorders in Parkinson's disease? *Pathophysiology* **11**(4) 187-196.

Moore DJ, West AB, Dawson VL and Dawson TM (2005). Molecular pathophysiology of Parkinson's disease. *Annual Review of Neuroscience* **28** 57-87.

Paliwal M and Kumar UA (2009). Neural networks and statistical techniques: A review of applications. *Expert Systems with Applications* **36**(1) 2-17.

Priyadarshi A, Khuder SA, Schaub EA and Priyadarshi SS (2001). Environmental risk factors and Parkinson's disease: a metaanalysis. *Environmental Research* **86**(2) 122-127.

Przedborski S, Vila M and Jackson-Lewis V (2003). Neurodegeneration: what is it and where are we? *Journal of Clinical Investigation* **111**(1) 3-10.

Rao V and Rao H (1996). C++ neural networks and fuzzy Logic, 2ⁿᵈ edition New Delhi India: *BPB Publication* 123-176.

Savitt JM, Dawson VL and Dawson TM (2006). Diagnosis and treatment of Parkinson disease: molecules to medicine. *Journal of Clinical Investigation* **116**(7) 1744-1754.

Sinha RK (2008). Artificial neural network and wavelet based automated detection of sleep spindles, REM sleep and wake states. *Journal of Medical Systems* **32**(4) 291-299.

Smith WW, Pei Z, Jiang H, Moore DJ, Liang Y, West AB, Dawson VL, Dawson TM and Ross CA (2005). Leucine-rich repeat kinase 2 (LRRK2) interacts with parkin, and mutant LRRK2 induces neuronal degeneration. *Proceedings of the National Academy of Science of the United States of America* **102**(51) 18676-18681.

Sulzer D (2011). How addictive drugs disrupt presynaptic dopamine neurotransmission. *Neuron* **69**(4) 628-649.

Takahashi R and Imai Y (2003). Pael receptor, endoplasmic reticulum stress, and Parkinson's disease. *Journal of Neurology* **250** (Suppl 3), III25-III29.

Tansey MG, McCoy MK and Frank-Cannon TC (2007). Neuroinflammatory mechanisms in Parkinson's disease: potential environmental triggers, pathways, and targets for early therapeutic intervention. *Experimental Neurology* **208**(1) 1-25.

Thomas KJ, McCoy MK, Blackinton J, Beilina A, Brug M, Sandebring A, Miller D, Maric D, Cedazo-Minguez A and Cookson M (2011). DJ-1 acts in parallel to the PINK1/parkin pathway to control mitochondrial function and autophagy. *Human Molecular Genetics* **20**(1) 40-50.

Research Article

Uversky VN, Li J and Fink AL (2001). Metal-triggered structural transformations, aggregation, and fibrillation of human alpha-synuclein. A possible molecular NK between Parkinson's disease and heavy metal exposure. *Journal of Biological Chemistry* **276**(47) 44284-44296.

Willis AW, Evanoff BA, Lian M, Galarza A, Wegrzyn A, Schootman M and Racette BA (2010). Metal emissions and urban incident Parkinson disease: a community health study of Medicare beneficiaries by using geographic information systems. *American Journal of Epidemiology* **172**(12) 1357-1363.

Yao Z, Gandhi S, Burchell VS, Plun-Favreau H, Wood NW and Abramov AY (2011). Cell metabolism affects selective vulnerability in PINK1-associated Parkinson's disease. *Journal of Cell Science* **124**(24) 4194-4202.