

**The use of multifocal visual evoked potential objective
perimetry for diagnosing optic neuritis primarily associated
with Multiple Sclerosis.**

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A thesis submitted to the Discipline of Clinical Ophthalmology and Save Sight Institute,
Faculty of Medicine, The University of Sydney, in fulfilment of the requirements for the
degree of Master of Medicine.

January 2006

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Declaration

I hereby declare that this thesis is my own original work, except where acknowledgment is made below and where due reference is made in the text. To the best of my knowledge and belief, this thesis does not contain material which has been accepted for the award of any other degree or diploma of a University or other institute for higher learning. All the experiments included in the thesis were performed in the Save Sight Institute and St Vincent's Hospital between January 2004 and November 2005.

I consent to the thesis being made available for photocopying and loan if accepted for the award of the degree.

Clare Fraser

January 11, 2006.

Acknowledgements

I would like to thank my supervisors Dr John Grigg and Dr Stuart Graham for their guidance and encouragement, particularly during the early stages when I was still in Perth. There are several other key people I would also like to thank: Dr Alex Klistorner though not an official supervisor has been equally involved on a day to day basis and has been of immense help; Dr Raymond Garrick who provided the patients, a VEP clinic and all the neurological advice and Professor Francis Billson who provided structural guidance and advice whenever it was needed. For their help in the statistical analysis of the data I would like to thank Richard Stump and Thomas John Naduvilath.

Finally, I want to dedicate this work to my grandmother, who was diagnosed with Multiple Sclerosis just after I was born.

Abbreviations used frequently

ASI – Accumap™ severity index

cVEP – conventional visual evoked potential (full field, pattern reversal)

LP – light perception

MRI – magnetic resonance imaging

MS – Multiple Sclerosis

mV - millivolts

mVEP – multifocal visual evoked potential

NAION – non arteritic ischemic optic neuropathy

NF 1 / 2 – Neurofibromatosis Type 1 or Type 2

NPL – non perception of light

ON – optic neuritis

ONSM – optic nerve sheath meningioma

ONTT – Optic Neuritis Treatment Trial

PPMS – primary progressive Multiple Sclerosis

ROC – receiver operator characteristics

RRMS – relapsing remitting Multiple Sclerosis

SPMS – secondary progressive Multiple Sclerosis

VEP – visual evoked potential

Summary

This thesis explores the capacity of multifocal visual evoked potentials to provide detailed information of optic nerve function. The aim is to determine particular ways in which optic neuritis and Multiple Sclerosis (MS) impact on the visual pathways by analysing the most appropriate use of amplitude and latency analysis. Knowledge of this impact will then allow analysis of the diagnostic capabilities of the multifocal visual evoked potentials in patients presenting with optic neuritis and possibly to predict their risk of progression to MS. In this way the thesis will be expanding the role of the mVEP into the field of neuro-ophthalmology, to fulfil its potential as more than a technique for objective perimetry.

Publications arising from this work

Fraser, C; Klistorner, A; Graham, S; Garrick, R; Billson, F; Grigg, J. *Multifocal visual evoked potential analysis of demyelinating or inflammatory optic neuritis*. **Ophthalmology**, 2006, 113 (2): p 323-333.

Fraser, C; Klistorner, A; Graham, S; Garrick, R; Billson, F; Grigg, J. *Multifocal visual evoked potential latency analysis: predicting progression to Multiple Sclerosis*. **Archives of Neurology**, 2006, 63: p 847-850.

Fraser, C; Klistorner, A; Graham, S; Garrick, R; Billson, F; Grigg, J. *Using neural network analysis of multifocal visual evoked potential trace arrays to improve diagnostic accuracy*. **Documenta ophthalmologica**, under first revision.

Fraser, C; Klistorner, A; Graham, S; Garrick, R; Billson, F; Grigg, J. *Comparison of multifocal visual evoked potentials with routine visual assessment in optic neuritis*. **Clinical and experimental ophthalmology**, submitted.

Fraser, C; Klistorner, A; Graham, S; Garrick, R; Billson, F; Grigg, J. *Multifocal visual evoked potentials in the differential diagnosis of optic neuritis*. Presented at **North American Neuro-Ophthalmology Society Meeting**, Copper Mountain, USA, February 2005. Winner "Best abstract by resident".

Aims

The first aim of this project was to determine if the multifocal visual evoked potential (mVEP) technique could reliably detect the presence of optic neuritis. The results would need to be sensitive and specific, and show some relation to currently available visual tests.

The study was designed to document the range of mVEP results seen in patients with a past history of optic neuritis, and to relate these results to the underlying pathology, in particular Multiple Sclerosis.

The study would explore which were the best mVEP measurement parameters, based on reproducibility and clinical significance for use in detecting optic neuritis, and potentially monitoring the course of the disease process. Techniques to optimize signal recognition and improve reproducibility using computerised neural networks would be examined.

The study would then prospectively use the mVEP system to track the changes in acute optic neuritis, from initial onset through to recovery. Applications of the mVEP in Multiple Sclerosis diagnostics for patients with optic neuritis would be examined.

Rationale

Conventional VEPs are known to show delays in demyelinating disease. However, research into the use of visual evoked potentials in the diagnosis of Multiple Sclerosis has decreased since the advent of magnetic resonance imaging (MRI) scans which have greatly advanced the area of MS diagnostics. The research presented here is important because it allows us to begin to utilise the emergent multifocal technology to revisit the use of VEP testing in a more advanced format. This new technique is objective and easy to perform, and should it prove useful in optic neuritis could be of great benefit to the patients and their clinicians. By providing much more information than the

conventional VEP, the multifocal VEP, may be able to bridge the gap between tests easily performed in a routine clinic and the diagnostic capabilities of the MRI.

Even after having MRI scans performed many patients are left with the uncertain diagnosis of “possible-MS” following an episode of optic neuritis. If the mVEP is able to provide further diagnostic information to the patient and clinician, and thus allow earlier diagnosis then it represents a worthwhile area of research. Though the validity of earlier diagnosis for a disease in which there is no cure may be questioned, recent research has shown that by commencing immune modulating therapy early in the course of MS, relapse rates and progression to disability can be reduced [1]. In addition, 90% of patients in one study became less anxious after being given a definitive diagnosis and expressed favourable feelings even though they faced a lifetime of chronic disease [2].

The mVEP may therefore provide a new diagnostic tool for the clinician in the assessment and management of the patient with optic neuritis.

Thesis outline

Chapter 1 - Introduction

The introduction describes both optic neuritis and Multiple Sclerosis (MS), in terms of their diagnosis, risk factors and significance in the community. Currently available means for assessing the visual pathways in optic neuritis and their limitations are discussed, along with current diagnostic techniques for MS. The role of conventional visual evoked potentials in the diagnosis of optic neuritis and Multiple Sclerosis is explored in detail. Some background to the multifocal visual evoked potential (mVEP) technique is given, along with detail of areas where it could be of potential benefit in optic neuritis and Multiple Sclerosis.

Chapter 2 – General methods

The General Methods section outlines the methods used in subsequent chapters. Patient enrolment and exclusion criteria are outlined for both the cross-sectional and longitudinal studies, as are the routine measurements taken during patient visits. The methods for assessing the visual function are described. The technique for performing mVEP testing and analysis are also described.

Chapter 3 – Sensitivity and specificity of mVEP, comparison with other tests

In this study, the results for the cross-sectional cohort are compared with normal controls, and patients with other causes for visual disturbance, in order to determine sensitivity and specificity of the mVEP in optic neuritis. The mVEP results are also compared to routine visual assessments in optic neuritis patients. This study showed that the mVEP was a viable method for assessing optic neuritis, as it had good sensitivity, specificity and corresponded with other indicators of visual dysfunction.

Chapter 4 – Reproducibility of mVEP

In order for the mVEP to be of clinical use in assessing optic neuritis or Multiple Sclerosis the test needs to be reproducible. Latency variability of the normal control group was calculated. Sectoral latency analysis was determined to be the most

reproducible method for latency assessment. Reproducibility was also determined for patients with known central scotomas due to optic neuritis.

Chapter 5 – Correlation of mVEP findings in optic neuritis and MS risk assessment

The latency and amplitude results for the cross-sectional group of patients were correlated with the Multiple Sclerosis category for each patient as per the McDonald Criteria for the diagnosis of MS. Latency values proved to be significantly different between each MS classification group. A one year follow-up of a subgroup of these patients proved that delayed latency test results may confer an increased risk of conversion to MS.

Chapter 6 – Acute optic neuritis mVEP results from initial onset to one year

The longitudinal group of acute optic neuritis patients underwent mVEP testing at regular intervals in order to determine if the technique can track changes within the optic pathways. Results were analysed in relation to the patient's MS classification

Chapter 7 – Improving mVEP trace recognition

Any technique used to monitor disease progress over time is limited by the retest variability. Artificial neural networks can be used in the classification of information. In this study an artificial neural network was designed to correctly identify true mVEP signals from background EEG noise and contamination. This could potentially improve the reproducibility of the mVEP, increasing diagnostic certainty.

Chapter 8 – Conclusion

This section provides a summary of all the study findings, and how the results interlink, providing an overview of how the use of the mVEP in optic neuritis and Multiple Sclerosis has been established as a potential diagnostic technique.

Chapter 1

Review of the literature

A Optic neuritis and Multiple Sclerosis

i. Description and Demographics

Definition

Optic neuritis (ON) is a term that describes an optic nerve pathology due to idiopathic, inflammatory, infectious or demyelinating causes that is usually acute and unilateral. If there is optic nerve swelling on fundoscopic examination, then the term *papillitis* or *anterior optic neuritis* can be used. Papillitis may be associated with flame shaped haemorrhages and is the most common type of optic neuritis in children. If the optic nerve appears normal on examination, then the optic neuritis is classified as *retrobulbar optic neuritis* and is the most frequent type seen in adults. *Neuroretinitis* is the least common type of ON, and is usually associated with a viral infection or cat-scratch fever. On examination there is papillitis in conjunction with a macular star composed of hard exudates [3]. Most ophthalmologists use the term optic neuritis to describe idiopathic or demyelinating ON [4].

Diagnosis

Optic neuritis affects otherwise healthy young individuals, usually aged 20-30 years. Women are affected two to three times more commonly than men. ON causes a decrease in vision over a 7-10 day period, and is commonly associated with pain on eye movement [5]. The majority of patients with a painful ON have involvement of the orbital segment of the optic nerve. The absence of pain suggests the ON is limited to the canalicular or intracranial portion of the optic nerve [6]. Patients with a typical presentation of idiopathic or demyelinating ON have the features seen in Table 1.1. These were the

clinical characteristics seen in the 455 patients enrolled in the Optic Neuritis Treatment Trial (ONTT) between 1988 and 1991 [7].

Differential diagnosis

The features listed in Table 1.2 reflect the “typical” ON presentation. Those patients presenting with atypical features, should be examined further. Atypical features include: severe headache, uveitis, retinal inflammation, failure for vision to improve after 30 days, over 50 years of age and evidence of another systemic condition. Full history and examination should guide further investigations such as syphilis serology and antinuclear antibodies [7]. The presence of associated polyneuropathies should lead to consideration of Guillain-Barre or Miller-Fisher syndrome[4]. Patients should also be asked about alcohol consumption, tobacco use and other drugs such as ethambutol, isoniazid and amiodarone – all of which are known to cause optic neuropathy [5]. The common causes of atypical optic neuritis and the mimics of optic neuritis are presented in Table 2.

In children optic neuritis may be para-infectious, associated with measles, mumps, chickenpox, glandular fever or following immunisation. In these cases patients usually present 1-3 weeks following the infection with severe, often bilateral, visual loss and papillitis [3].

Characteristic	Patients
Female	77%
Caucasian	85%
Age (years)	32 +/- 6.7
Ocular pain	92%
Pain worse on eye movement	87%
Optic disc	
Normal (retrobulbar)	65%
Swollen	35%
Mild diffuse swelling	51%
Mild focal swelling	29%
No retinal or optic disc haemorrhage	84.5%
Normal vitreous	94%
Visual acuity	
20/20 or better (6/6)	11%
20/25 – 20/40 (6/7.5 – 6/12)	25%
20/50 – 20/190 (6/15 – 6/57)	29%
20/200 – 20/800 (6/60 – 6/240)	20%
Count fingers or hand motions	10%
Light perception	3%
No perception of light (NPL)	3%
Visual field defect of affected eye	
Diffuse	48%
Altitudinal, arcuate, nasal step	20%
Retrochiasmal	9%
Central, cecocentral	8%
Chiasmal	5%
Other	24%
Abnormal MRI (one or more significant white matter lesion)	49%

Table 1.1: Clinical profile of the Optic Neuritis Treatment Trial patients [7].

In the elderly population other causes of visual loss must be considered before a diagnosis of optic neuritis is made. Non-arteritic ischemic optic neuropathy (NAION) may present after the age of 45 with a sudden, painless visual loss and typically an altitudinal visual field defect. Giant cell arteritis usually presents in those aged 60 to 80 and is typically associated with: scalp tenderness, headache, jaw claudication, polymyalgia rheumatica and malaise [5].

The association of acute or subacute loss of vision in one eye or both eyes caused by optic neuropathy with a simultaneous or separate attack of transverse or ascending myelopathy is referred to as neuromyelitis optica (Devic's disease). The disease can be monophasic or relapsing, with a high relapse rate in the first two years of disease associated with a poor prognosis and often occurs in the Asian population [8].

Retinal disease	
Compressive optic neuropathy	Tumour Idiopathic intracranial hypertension
Toxic optic neuropathy [5]	Alcohol-tobacco amblyopia (Vit B def) Amiodarone Ethambutol
Genetic disease [9]	Leber's hereditary optic neuropathy Autosomal dominant ON (Kjer syndrome) Wolfram Syndrome (DIDMOAD= diabetes insipidus/mellitus, optic atrophy, deafness)
Infections	Syphilitic ON (primary or secondary) Human immunodeficiency virus (HIV) Cat-scratch (Bartonella Henselae) Tuberculosis Post-viral Aspergillus
Inflammatory	Sarcoidosis Systemic lupus erythematosus (SLE) Reiter's syndrome
Vasculitis	Giant cell arteritis (temporal arteritis) Polyarteritis nodosa Wegner's granulomatosis Churg-Strauss syndrome
Paraneoplastic optic neuropathy [10]	Typically lung cancer, CRMP-5 IgG +ve

Table 1.2: Summary of differential diagnoses in atypical optic neuritis[11]

Visual prognosis for typical optic neuritis

The course of visual recovery in typical optic neuritis is often rapid regardless of treatment, with improvement noticeable within the first two weeks in most patients and much of the recovery occurring by the end of one month [4]. Over 75% of patients recover to normal vision within 6 months [12]. If recovery of vision is incomplete at 6 months, some further improvement may continue for up to one year. Ten years after optic neuritis in the ONTT, 70% of patients had 20/20 acuity in both eyes and 86% had 20/20 vision in one eye. Relapses of optic neuritis within this time did not appear to cause a major loss of visual function [13]. However, patients may complain of residual deficits in colour vision, stereopsis and light brightness perception, despite the apparent normalisation of distance vision [14].

The only predictor of poor visual outcome found in the ONTT, was poor visual acuity at the time of entry to the study. However, of the patients with visual acuity of light perception (LP) and non perception of light (NPL), 67% recovered to a final visual acuity of 20/40 or better. Older age at onset was statistically associated with slightly worse visual outcome [13].

Association with Multiple sclerosis

The incidence of ON within the US is 3 per 100 000 people [15]. Despite this relatively low incidence and the fact that vision recovers in the majority, optic neuritis is still of great significance due to its association with Multiple Sclerosis (MS) [16, 17]. The risk for developing MS following ON is quite variable within the literature, reflecting differences in the populations studied, however the majority of studies indicate a 25-35% risk [18]. In fact the most common cause of visual loss in MS patients is optic neuritis, with 90% of MS sufferers experiencing some form of ON during the course of their disease [19].

Following the release of the 10 year data from the Optic Neuritis Treatment Trial (ONTT) it is now known that a single magnetic resonance imaging (MRI) white matter lesion in the brain of greater than or equal to 3 millimetres increases the risk of future MS. In those

ON patients with a normal brain scan the absence of pain on eye movement, no perception of light vision, severe optic disc swelling, haemorrhages and exudates are associated with a low risk of future MS [20].

The disease now known as Multiple Sclerosis was first described in 1868 by Jean-Martin Charcot as “sclerose en plaque”[21]. Multiple Sclerosis is characterised by episodic neurological dysfunction caused by localised damage to nerve fibres within the brain and spinal cord. A typical MS lesion causes the loss of myelin. This demyelination, results in a slowing or complete blockage of the neuronal signals, thus resulting in the neurological dysfunction. Early in the disease course remyelination occurs and the neurological function will often recover to some extent. This phenomenon was thought to be due to the preservation of the nerve fibres themselves. It has now been demonstrated that this process of demyelination causes damage to the underlying nerve axons, and ultimately leads to brain atrophy and the accrual of progressive neurological dysfunction [22]. Once the degenerative process starts, the changes are irreversible, thus treatment is most effective when started early. For this reason there is an imperative to diagnose MS as early as possible.

The overall risk factors for Multiple Sclerosis in an individual are also well documented. Caucasians of northern European heritage are the most commonly affected ethnic group, with women two to three times more frequently affected than men. Living in a temperate climate causes a higher risk of MS, and those who migrate from a high risk to a low risk area after the age of 15, remain at high risk [1]. Several infectious agents have been postulated to be the environmental trigger for MS, with the current leading candidates being Epstein-Barr, Herpes VI and Chlamydia pneumoniae [23]. Other recent work has highlighted an increasing incidence of MS in previously low risk areas such as Queensland and California. With low levels of Vitamin D now thought to contribute to the onset of MS, it appears that the increased used of sunscreen and decreased sun exposure in these areas may be contributing to a rise in the incidence of MS [24]. A genetic predisposition to MS has been identified, though the only significantly associated genotypes are the major histocompatibility complexes (HLA) DR and DQ [25].

The National Health Survey of Australia in 2001 indicated that approximately 15 000 Australians had a diagnosis of MS, the figure is now thought to be closer to 22 000, and world wide the figure is close to 2.5 million. The MS Society of New South Wales (Australia) estimates that 230 people were diagnosed with MS in this state alone in the last 12 months [26]. As MS, like ON, is typically diagnosed in the third and fourth decades, it results in significant functional and often work-related disability, in what should be the most productive years of life. A recent media release estimated (by aggregating personal and community costs associated with diagnosing, treating and living with the disease) that MS costs \$2 billion per year in Australia alone [27].

ii. Classification of MS

The key issue in diagnosing MS is to provide evidence for dissemination of demyelinating lesions in both time and space within the central nervous system. In the past this was based solely on clinical observations, but more recently magnetic resonance imaging (MRI) standards have been incorporated into the diagnostic criteria.

Over the last 40 years there have been 3 main diagnostic criteria used by neurologists to make the diagnosis of MS, these are described below.

Schumacher criteria

The Schumacher criteria were developed in 1965 [28, 29]. The criteria are as follows:

- Neurological examination reveals objective abnormalities of central nervous system (CNS) function
- History indicates involvement of two or more parts of the CNS

Involvement of CNS follows one of two patterns:

- Two or more episodes, each lasting at least 24 hours and at least one month apart

- Slow or stepwise progression of signs and symptoms over at least 6 months
- Patient aged 10-50 at onset
- Signs and symptoms cannot be better explained by another disease process

There were no specific allowances in the criteria for diagnostic tests, the diagnosis essentially being a clinical one.

These criteria led to the following designations:

- Clinically definite MS – which could be made if all the Schumacher criteria are fulfilled
- Probable MS – which refers to relapsing/remitting MS (RRMS) symptoms where only one neurological symptom commonly associated with MS is found or if there is only a single attack and there was no better explanation for the symptoms
- Possible MS – this refers to RRMS without documented signs or where the objective signs are insufficient to establish more than one site of CNS involvement.

Poser criteria

The Poser criteria were proposed in 1983 as an update to the Schumacher criteria, are still used by some neurologists. They were developed to reflect the advances of detection techniques such as MRI, VEP and cerebrospinal fluid (CSF) analysis. These techniques allowed neurologists to determine the existence of clinically silent lesions. The criteria are as follows:

- Clinically definite MS
 - 2 attacks and clinical evidence of 2 separate lesions
 - 2 attacks, clinical evidence of one and paraclinical evidence of another separate lesion
- Laboratory supported definite MS
 - 2 attacks, either clinical or paraclinical evidence of 1 lesion, and CSF immunological abnormalities

- 1 attack, clinical evidence of 2 separate lesions and CSF abnormalities
- 1 attack, clinical evidence of 1 and paraclinical evidence of another separate lesion, and CSF abnormalities
- Clinically probable MS
 - 2 attacks and clinical evidence of 1 lesion
 - 1 attack and clinical evidence of 2 separate lesions
 - 1 attack, clinical evidence of 1 lesion, and paraclinical evidence of another separate lesion
- Laboratory supported probable MS
 - 2 attacks and CSF abnormalities

McDonald criteria

In July 2000, the International Panel on the Diagnosis of MS updated the diagnostic criteria for MS to integrate MRI findings into the overall diagnostic scheme. The diagnostic criteria are presented as clinical pathways, firstly the clinical presentation is given, followed by the additional investigational data required to make a diagnosis of MS. Table 1.3. Patients not fitting these criteria are given a diagnosis of “Not-MS”, while those who display some but not all features required for diagnosis are labelled “possible MS”. The panel agreed to follow the Barkof criteria for MRI diagnosis of MS. Table 1.4. This classification relies on the traditional need to demonstrate dissemination of clinical events and lesions in time and space, as well as requiring objective clinical evidence for these events [30]. These criteria make it possible to diagnose MS more accurately and earlier in the disease course.

Table 1.3: MacDonald classification requirements for a diagnosis of MS. If the patient has one of the following clinical presentations then they must also fulfil the extra requirements listed in the corresponding column on the right hand side of the table.

Clinical presentation	Extra data for a diagnosis of MS
<ul style="list-style-type: none"> • 2 or more attacks (relapses) • 2 or more objective clinical lesions 	None; clinical evidence will suffice (additional evidence must be consistent with MS)
<ul style="list-style-type: none"> • 2 or more attacks • 1 objective clinical lesion 	Dissemination in space, demonstrated by: <ul style="list-style-type: none"> • MRI OR • 2+ MRI lesions consistent with MS plus positive CSF. OR • A further clinical attack involving a different site
<ul style="list-style-type: none"> • 1 attack • 2 or more objective clinical lesions 	Dissemination in time, demonstrated by: <ul style="list-style-type: none"> • MRI OR • A second clinical attack
<ul style="list-style-type: none"> • 1 attack • 1 objective clinical lesion (monosymptomatic presentation)	Dissemination in space demonstrated by: <ul style="list-style-type: none"> • MRI OR • Positive CSF and 2 or more MRI lesions consistent with MS AND Dissemination in time demonstrated by: <ul style="list-style-type: none"> • MRI OR • A second clinical attack
<ul style="list-style-type: none"> • Insidious neurological progression suggestive of MS (primary progressive MS)	<ul style="list-style-type: none"> • Positive CSF AND Dissemination in space demonstrated by: <ul style="list-style-type: none"> • MRI evidence of 9 or more T2 brain lesions OR • >2 spinal cord lesions OR

	<ul style="list-style-type: none"> • 4-8 brain lesions and 1 spinal lesion OR • positive VEP with 4-8 MRI lesions OR • positive VEP with <4 brain lesions plus 1 spinal cord lesion <p>AND</p> <p>Dissemination in time demonstrated by:</p> <ul style="list-style-type: none"> • MRI. OR • Continued progression over one year
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Table 1.4: Barkof criteria for MRI abnormality consistent with MS.

Three of four of the following:

1. One gadolinium enhancing lesion or 9 T2-hyperintense lesions if there is no gadolinium enhancement.
2. At least one infratentorial lesion.
3. At least one juxtacortical lesion.
4. At least three periventricular lesions.

Dissemination in time can be evidenced on MRI by either a gadolinium enhancing lesion demonstrated in a scan done at least 3 months following onset of clinical attack at a different site or a follow-up scan after an additional 3 months showing a gadolinium enhancing lesion or a new T2 lesion. Barkof does not include spinal cord plaques in the criteria, but states that they are as relevant as brainstem plaques (classified as infratentorial) [31]

An “attack” is classified as a neurological disturbance of the kind seen in MS, with a minimum of 24 hours duration, as a subjective report or objective observation. Attacks are considered to be separate if there is a minimum of 30 days between the onset of event 1 and the onset of event 2.

Abnormality in paraclinical tests is also defined. For abnormal CSF, there must be oligoclonal bands in the CSF but not the serum. Evoked potentials are considered abnormal if they are delayed but the waveform is still well preserved.

These criteria have been extensively used and tested since their introduction in 2001. Studies have shown them to be both more sensitive and specific than the Poser criteria, resulting in earlier diagnosis of MS. Modifications have been made by an international panel in 2005, which set out to simplify the process required to demonstrate dissemination of lesions in time and clarify the use of spinal cord lesions in the diagnosis [32, 33].

iii. Treatments available

It had been recognised that treatment with corticosteroids in the setting of optic neuritis resulted in a more rapid recovery of visual function. However it was also reported that there was no difference in visual outcome by 6-12 months [34]. The Optic Neuritis Treatment Trial (ONTT) set out to provide the definitive data to evaluate the efficacy of corticosteroid treatment for acute optic neuritis. The trial enrolled 457 patients with a relative afferent papillary defect as well as a visual field defect in the affected eye. Other causes for optic neuritis were ruled out prior to entry in the study. The patients were randomly assigned to one of three treatments:

1. Intravenous (IV) methylprednisolone (250mg 4 times daily for 3 days) with an 11 day oral steroid taper.
2. Oral prednisolone (1mg/ kg once daily) for 14 days
3. Oral placebo for 14 days

The study confirmed that IV corticosteroids with an oral taper resulted in an acceleration of visual recovery, however there were no long term benefits to visual outcome. Interestingly, it was also found that the first dosing regime reduced the rates of patient

conversion to Multiple Sclerosis over the first 2 years post ON. However this effect was not sustained into the third year post diagnosis [18].

Pathological and MRI studies now suggest that axonal damage is an early event in the evolution of MS, and that this is primarily dependent on inflammatory processes [22]. Treatments for MS available include interferon beta, which has a partly anti-inflammatory action.

The Controlled High Risk Subjects Avonex Multiple Sclerosis Prevention Study (CHAMPS) was a randomised, double blind, placebo controlled trial in patients who had a first acute demyelinating event (optic neuritis, incomplete transverse myelitis, brain-stem or cerebellar syndrome) and evidence of previous subclinical demyelination on MRI. Eligible patients had 2 or more clinically silent brain lesions of 3mm or more in diameter on MRI, one of which was ovoid or periventricular. Patients were randomised to receive either 30 micrograms of intra-muscular interferon-beta-1a (Avonex) or placebo weekly. Results indicated that treatment with interferon significantly reduced the risk of developing MS, according to the Poser criteria, by 44% compared with placebo [35]. In a more recent analysis of the CHAMPS data, it was found that using more stringent MRI criteria for eligibility as set out by Barkof, being 9 or more white matter lesions with at least one gadolinium enhancing lesion, that treatment with interferon conferred a 66% reduction in risk for MS over 3 years [36].

The more recent Betaferon in Newly Emerging MS for Initial Treatment (BENEFIT) study used interferon beta-1b, in a double-blind, placebo-controlled randomised trial in patients with a first episode suggestive of MS. Patients were followed over time, with conversion to MS defined according to the McDonald criteria. Results show that the risk of developing MS within a year was reduced by 46% when compared to the placebo group [37]

A review of all available preparations of interferon and the available studies on each, concluded that treatment with all interferon-beta products is effective compared to

placebo, with a possible advantage to a three times weekly dosing regimen, rather than weekly [38].

Interferon-beta-1a has been approved for use in Europe and America for the use in patients who have experienced a first clinical episode and have MRI features consistent with MS (optic neuritis, incomplete transverse myelitis, brain-stem or cerebellar syndrome), where alternative diagnoses have been excluded and the patient is deemed to be at high risk for developing MS. This treatment option is not yet available in Australia. However, given that there is now a treatment to reduce the risk of MS in patients post optic neuritis, the need for objective tests which may reflect a higher risk for progression to MS becomes apparent. Interferon therapy is not without side effects, the most common being flu-like symptoms and depression. Thus preventative therapy must be guided by this knowledge of which patients are inherently at a higher risk of MS.

B Methods for assessing optic pathways

i. Basic tests for assessing optic nerve function

The test for a relative afferent papillary defect (RAPD) is an important bedside test used to determine if decreased vision in a patient is due to an optic nerve problem. Studies suggest that RAPD is mainly related to the integrity of central vision being correlated with a reduction in conventional VEP amplitudes and intraocular mean deviation on Humphrey subjective perimetry in patients with optic neuritis [39, 40].

Loss of colour vision can be inherited or acquired. Though there are many causes of acquired loss of colour vision, optic neuritis causes colour vision abnormalities in 88% of affected patients [7]. The ONTT patients showed mixed red-green (RG) and blue-yellow (BY) colour defects. BY defects tend to be more common in the acute phase of the disease, with slightly more RG defects at 6 months. It was thus concluded that the type of

colour defect cannot be used in the differential diagnosis of optic neuritis [41]. Red desaturation can also be measured using a red test object.

Brightness perception can be tested using a pen light. Poor brightness perception is reported in 89% of patients with optic neuritis and can be used to follow the course of disease [42, 43]

Contrast sensitivity is the ability of the eye to distinguish subtle degrees of contrast. Retinal, optic nerve disease and clouding of the ocular media can impair this ability. In patients with optic neuritis but normal visual acuity, contrast sensitivity abnormality rates have been reported at 99% [44].

ii. Visual field and perimetric analysis

The visual pathways extend from the retina to the occipital pole, encompassing an extra-cerebral course, as well as the hemispheric white matter. Lesions affecting the nerve fibres along these pathways cause a variety of visual field defects. Due to the complex yet predictable course of the nerve fibres within the visual pathways, any lesion to the fibres at a certain location will produce equally predictable visual field defects, see Figure 1.1.

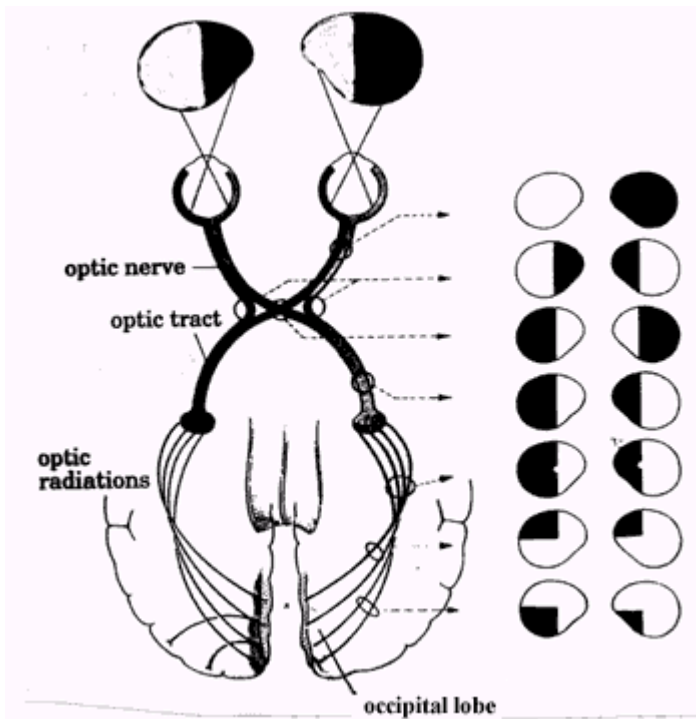


Figure 1.1: Lesions along the optic pathways create predictable visual field defects.[3]

Visual field testing can be divided into kinetic or static testing which can be manual or automated. Automated perimetry though lacking in defect shape detail due to the six-degree stimulus area, proved to be more sensitive than kinetic testing. However, the Humphrey Visual Field Analyser (HVFA) sensitivity relies on a co-operative and competent patient, as well as a good technician to monitor the patient and despite the introduction of the Swedish Interactive Threshold Algorithm, which has reduced the test time by half, there is still high test-retest variability [45].

Patients with ON involved in the Optic Neuritis Treatment Trial (ONTT) underwent HVF testing. Initial perimetry revealed that 48% present with a diffuse field defect (see Figure 1.2), 20% with altitudinal or arcuate defects and 8% present with a centrocecal defect [46]. These results were initially surprising as they went against the previously held belief that ON caused a centrocecal scotoma. However it is now believed that the central scotoma found by Goldmann perimetry represents diffuse suppression of sensitivity within the central 30 degrees of vision [47]. Although patients with ON can present with central visual field loss and sparing of the periphery, it is rare for the peripheral field to be

abnormal in the presence of a normal central field [4]. In most cases of ON the recovery can therefore be tracked effectively using automated perimetry of the central visual field.

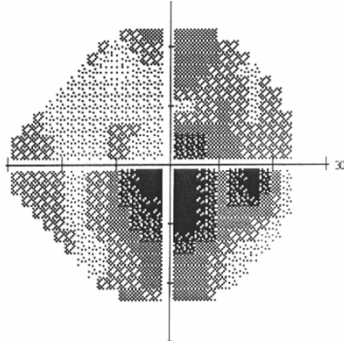


Figure 1.2: Humphrey visual field analysis, grey-scale map of a patient with optic neuritis, showing diffuse loss of sensitivity with a dense central scotoma.

iii. Visual Evoked Potentials

Visual evoked potentials represent a specific change in ongoing electroencephalographic (EEG) recording due to stimulation of the visual pathway to either a pattern or flash stimulus. Recordings of evoked potentials can be made with the use of electrodes applied to the scalp, with the main signal detected over the occipital cortex.

The VEP signal is mainly derived from the macular region as the predominant function of the occipital cortex is to subserve macular function. Therefore it has been estimated that 65% of the total VEP response represents the central 2 degrees of the visual field [48]. However studies were conducted using selective stimulus of central, nasal and temporal regions of the visual field to determine the retinotopic variation of conduction delay in ON [49] It has been reported that little or no VEP activity can be recorded beyond 15 degrees of eccentricity [50].

The evoked potential can be extracted from the underlying EEG using time-locked computer-averaging techniques, and reflect processing by the cortex and sub-cortex. This cortical activity relates to the physical characteristics of the stimulus applied [51].

The standard ISCEV protocol for VEP testing [52] recommends using a midline and two lateral active electrodes, to ensure that chiasmal and retrochiasmal lesions are not missed. Pattern-reversal VEP recordings are considered the least variable. The patient is sat one-meter from a checkerboard whose black and white squares alternate every 1-2 seconds. A spatial frequency of 10-20 min of arc with a temporal frequency of 8Hz gives the optimal response [48]. This results in a large slow biphasic wave being recorded from the averaged results of over approximately one hundred checkerboard reversals. The first positive component at approximately 50-70ms, called N75 is believed to reflect the activity of the striate cells in area 17. While the most prominent deflection of the wave is usually 100ms (95-110ms) after stimulus presentation, and is named the P100 (see Figure 1.3). This is thought to originate in the ventral extra-striate cortex of the fusiform gyrus [53]. The amplitude and latency of the P100 are measured and compared to normal population results and to the fellow eye. Prolongation of the P100 indicates delay primarily in the pregeniculate pathway [54].

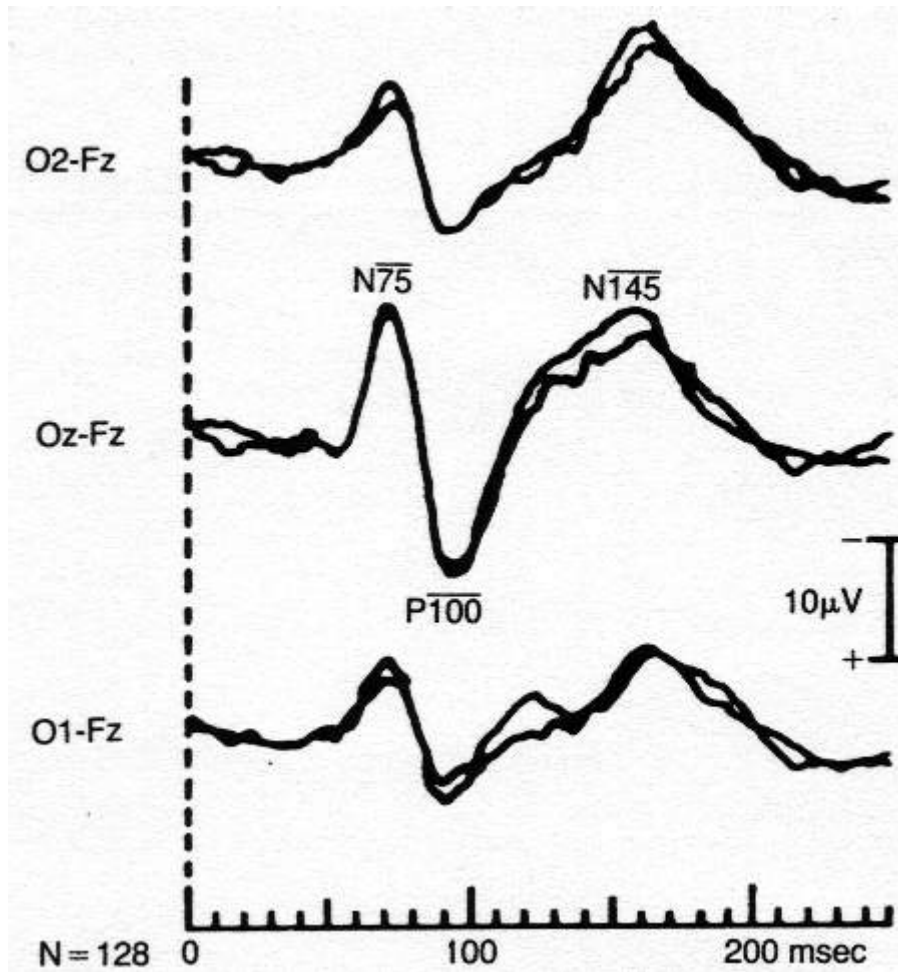


Figure 1.3: 3 lead conventional VEP results.

(Source: www.angelfire.com/retro/michaelpoon168/visual_evoked_potentials.htm)

The advent of visual evoked potentials (VEP) in the 1970s first allowed clinicians to assess neural conduction in the optic nerve. Halliday demonstrated that diagnostic confirmation of optic neuritis can be made using full-field conventional VEP delays [55-57]. The full role of VEP in optic neuritis will be described in detail later.

Optic neuritis can cause a loss of VEP signal amplitude as well as a delay in the signal. A loss of signal amplitude is also seen in compressive lesions of the optic nerve and latency

delay is found in Parkinson's disease and migraineurs. Overall the VEP has been shown to be very sensitive in these conditions however it is a non-specific diagnostic tool. [58]

C Diagnostic tests for Multiple Sclerosis

As the majority of disease in Multiple Sclerosis is asymptomatic, there is a need for surrogate markers of disease to evaluate many aspects of MS such as susceptibility, risk of developing MS after a first attack, disease activity, progression, prognosis and treatment response. [59] A number of markers are already in use and include magnetic resonance imaging (MRI), detection and quantification of auto-antibodies and cytokines. Clinical measures such as the degree of disability and the relapse rate are relatively insensitive to change and only weakly predictive of long-term clinical outcome. Despite this no laboratory test has been developed, including MRI, that meets the requirements of the FDA for a surrogate marker of disease prognosis [60].

i. Magnetic resonance imaging

MRI, being able to measure different components of disease activity and detect clinically silent MS lesions, is considered the best surrogate marker for MS disease activity available [61].

The basis of MRI is to generate contrast between tissues by altering the physical properties of their component atoms using a strong magnetic field and by measuring responses provide precise anatomical detail. A routine MRI can provide further information with the use of Gadolinium (Gd) as a contrast medium to enhance the MRI image. When acute inflammation of MS lesions causes the blood-brain-barrier to be damaged Gd can enter the lesion, allowing for differentiation between acute and chronic lesions. Another subset of MS lesions can be seen as hypointense areas, or "black holes" which indicate extensive tissue destruction and probably represent axonal loss and gliosis [59].

Periventricular white matter changes on MRI consistent with MS have been reported in 40-70% of patients with isolated optic neuritis [62, 63]. The use of Gd may increase the detection of disease activity [64]. From the results of the ONTT, brain MRI scans performed at entry to the study were a strong predictor of developing MS, with the 10-year risk of MS ranging from 22% in patients with no MRI lesions, to 56% in patients with one or more lesions [20].

However, several problems exist with MRI scanning. Firstly, cranial MRI scans are expensive to the patient and the community. Secondly, the rate of adverse events due to IV gadolinium, required for the diagnosis of “active” MS lesions, is reported to be between 1-2.9% [15]. Thirdly, the routine MRI gives only structural information, not functional information on the state of the visual pathways. In addition, it has been reported that while cranial MRI is a specific test for MS, the sensitivity and negative predictive value are not sufficiently high for a normal MRI to be used to conclusively exclude a diagnosis of MS [65]. There are several conditions that can leave the appearance of white matter lesions on MRI, such as autoimmune disorders and microvascular disease. Finally, absence of MRI abnormalities does not protect against the future development of MS [20].

Advanced MRI methods such as magnetization transfer, diffuse-weighted imaging [66] and functional MRI, while improving sensitivity, are limited by their cost, availability, complexity and have a low-to-modest correlation with disability [67]. MRI spectroscopy is used to measure the concentration of N-acetyl aspartate as a marker of functioning axons. The loss of N-acetyl aspartate correlates with disability, however, the studies have been small, and the prognostic value of this test is yet to be evaluated [68].

ii. Lumbar puncture

The biochemical analysis of the cerebrospinal fluid (CSF) is important in the diagnosis of MS. Increased immunoglobulin within the CSF of MS patients is common and the detection of oligoclonal bands is the most specific CSF test for MS. The presence of oligoclonal bands in the CSF but not the serum is generally considered to be crucial for diagnosis. However, oligoclonal bands are also found in other CNS conditions, such as acute disseminated encephalomyelitis (65%) and acute stroke (10%), probably reflecting cerebral inflammation [59].

Patients who present with Multiple Sclerosis at the time of their optic neuritis may show cerebrospinal fluid (CSF) abnormalities. The most common findings include an increased cell count (>5 cells/ mm^3), oligoclonal bands, increased CSF-immunoglobulin G (IgG) and increased total protein. However in the ONTT a lumbar puncture with CSF analysis did not increase the diagnostic rates for MS. A normal CSF analysis following an initial episode of ON, does not preclude an eventual diagnosis of MS [69]. Further analysis from the ONTT revealed that CSF analysis was useful in the MS risk assessment following optic neuritis, only when the MRI scan was normal. The presence or absence of oligoclonal bands in patients with abnormalities on MRI did not increase the positive predictive value. The final recommendation is that CSF analysis, being an invasive test with the known complications of headache, CSF leak and spinal abscess, should only be performed in cases of atypical optic neuritis or in cases where the diagnosis of MS might be clarified by the results [4].

D Current role of VEP in optic neuritis and Multiple Sclerosis

The current recommendation is that conventional VEP is often abnormal in patients with ON, and abnormal VEP in the setting of a clinically diagnosed ON does not alter the diagnostic or treatment plan [4]. Visual evoked potentials are only considered useful in

identifying a second site of neurological involvement to strengthen the clinical diagnosis of MS in patients with no history of an optic neuropathy [30].

Previous VEP studies on inflammatory ON and MS have often reported conflicting results, which to some extent may be explained by lack of consistency in the classification standards used. Some studies have divided the patients into acute mono-symptomatic optic neuritis (AMON) or optic neuritis as part of MS [57, 70]. Other studies have chosen the Poser criteria [17, 71-74], or studied possible MS versus probable MS combined with definite MS [75-78]. However, not all patients in these MS studies had a documented episode of optic neuritis. Thus, Mauguiere reported 80% abnormality in those with MS, compared to 50% abnormality rates in those with possible or probable MS [75]. Similar findings were also documented with abnormality levels in those with MS (with or without a history of optic neuritis) between 68-100%, and those with possible or probable MS between 50-70% [75-78]. No study reported a significant difference between the groups.

Studies reporting amplitude abnormalities range from 17% to 100% in MS alone [79, 80], and 78% to 100% in retro-bulbar ON alone [57]. When comparing amplitude differences between groups, either larger amplitudes in patients with MS versus those with AMON (at 2 months post ON only) [74], or no significant difference between groups [57, 81] was found.

The literature describing latency changes in optic neuritis patients does not give a complete picture regarding latency recovery in conventional VEPs. Halliday described that latency delays may persist for many years after acute optic neuritis [55, 56] and several other studies of both cross sectional [81, 82] and longitudinal design [83, 84] have shown long term latency reductions. Garrick et al found that patients either worsened (12%), recovered (38%) or the latency remained unchanged (50%) [85]. Brusa studied 31 patients, 22 with isolated optic neuritis and 9 with MS, no significant difference in latency analysis or visual function was found between the two groups [81]. However, 2 studies found that the mean latency was significantly shorter in patients with acute mono-

symptomatic optic neuritis (AMON) when compared to those with clinically definite MS at all testing times between acute onset and one year [57, 74].

Since the early 70's it has also been recognised that the waveform induced by pattern VEP can be bifid or W-shaped, rather than the single undivided deflection of P100. It was thought that this w-VEP was associated with demyelination, though it has been shown to be present in some normal studies [86]. Thus a long-term follow-up study of 97 patients was performed. Authors concluded that bifid VEP is rarely observed in healthy subjects, and the presence of a bifid wave was significantly more frequent in patients with demyelinating pathology ($p < 0.01$ Chi-squared) [87].

Despite an apparent declining interest in VEP since the advent of MRI scans much work has continued to find ways to improve the results. In cases where the MRI findings are minimal, or when MRI has lower specificity (i.e. older subjects), the conventional VEP can provide valuable diagnostic information.

In a recent study from the UK, the use of low contrast stimuli in addition to the standard high-contrast stimuli were used when testing patients with MS and ON [88]. This retrospective study of 124 patients used 95% and 20% contrast with 50' and 20' checks to measure a cVEP. It was found that low contrast small check sizes increased the number of abnormalities detected on the intra-ocular latency deficit.

Abnormal VEP in subjects with optic neuritis is reported between 65-100% [57, 73, 79, 84, 85, 89, 90]. Latency delays with a preserved waveform in full-field VEP have been shown to be a hallmark of established MS [91, 92]. However, it is thought that reliable prediction of the course of MS is not possible from VEP data [73]. Using alternate methods for measuring VEP, such as low contrast small checks, it was discovered that the apparent increase in intra-ocular latency deficits detected only represented an increased number of false positive tests, the test had no power to predict either ON or MS[88].

It is believed that, due to cortical magnification, full-field VEP signals are dominated by the central vision and do not provide the topographical detail of optic nerve damage caused by ON that has been seen with Humphrey subjective perimetry. However, contrary to classic teaching [93, 94], the Optic Neuritis Treatment Trial has shown that diffuse visual field defects are more common than the central and para-central defects which only occur in 8% of those with ON [95]. Thus conventional VEPs are unable to accurately record peripheral damage, and may overlook vital diagnostic and prognostic clues. In recent years there have been attempts to develop multifocal recording techniques that are capable of extracting focal VEP responses from more peripheral visual field locations, primarily as an objective means of detecting visual field scotomas (see below). This technology provides a unique opportunity to examine the cortical responses from multiple locations in patients with optic neuritis.

E The development of mVEP

i. An objective perimeter – first used in glaucoma

The perimetric tests described above (in section B) are all subjective and thus reproducibility is limited by the performance of the patient. Therefore there was a strong desire to develop an objective measure of visual function. Glaucoma causes nerve fibre loss producing a classic visual field defect [4]. The current gold standard for detection of this visual field loss is static automated perimetry. Detecting disease progression is a significant problem in glaucoma management. In particular determining at which point a change in perimetry indicates real disease progression requiring intervention rather than just an example of poor reproducibility [96]. Therefore glaucoma provided a good disease model in which to study objective perimetry.

Studies have shown that conventional VEP responses are abnormal in glaucoma, however sensitivity is not high and VEP cannot be used to monitor disease [97].

It has been shown that the spatial projection of retinal ganglion cells is maintained in the visual cortex, and thus a VEP from each representative area can be taken. The visual cortex is located on either side of the calcarine fissure in the occipital lobe. The cortical cells that subserve the axons from the peripheral fields lie anteriorly, with those from the macula at the extreme tip. The upper fields are represented in the lower half of the cortex, and the lower fields in the upper half [54]. This maintenance of proximity from the retina to the cortex means that as a particular segment of the retina is stimulated, it will cause a response in adjacent areas of cortex (see Figure 1.4). It is due to this maintenance of spatial proximity of fibres from the retina to projections in the cortex, that the multifocal technique could be considered.

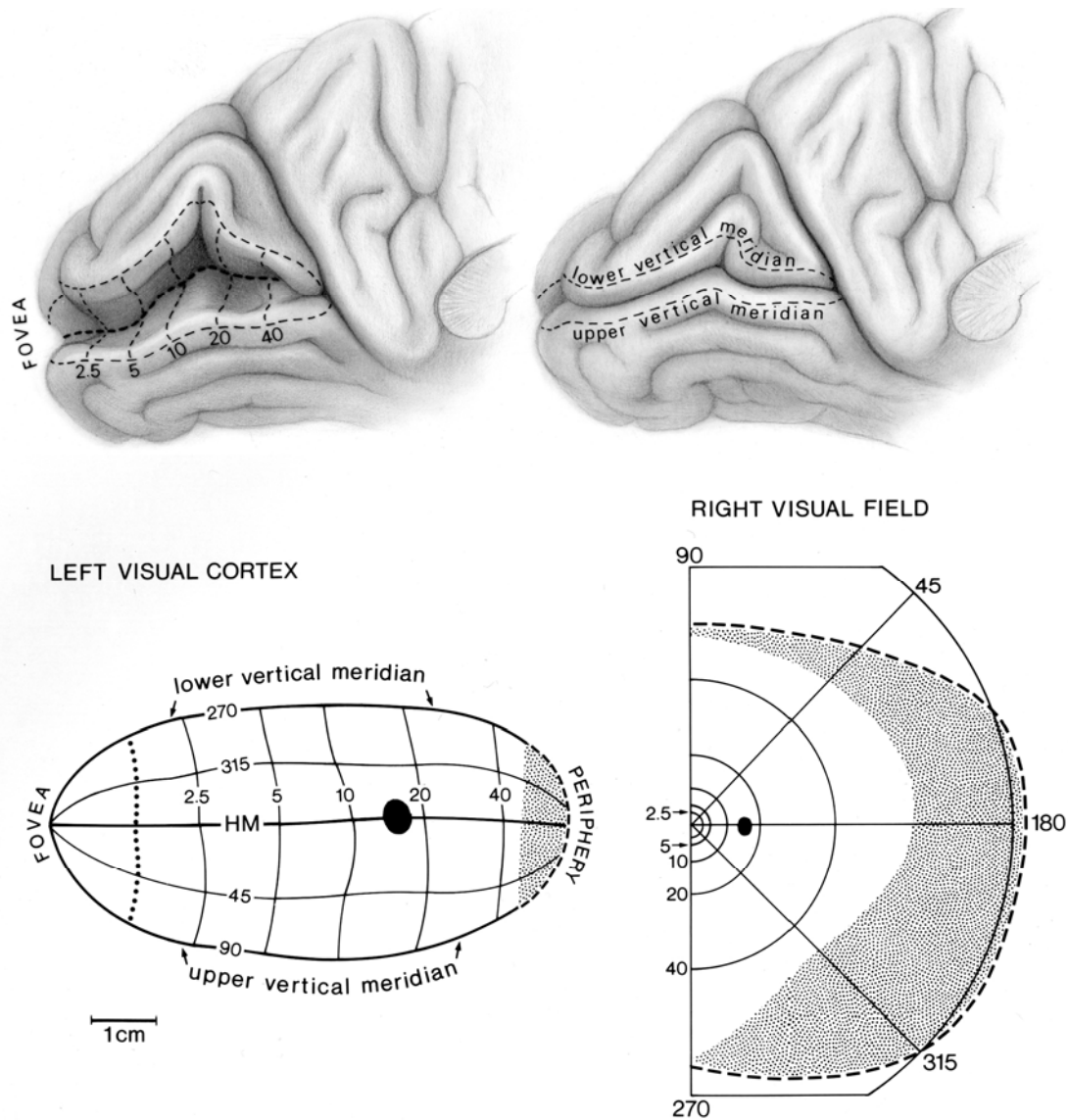


Figure 1.4: Maintenance of retinal topographic association in the visual cortex.

Reproduced from Horton et al, Archives of Ophthalmology, 1991 [98]

The new VEP technique developed by Baseler et al [99] allowed for the presentation of a multifocal stimulus, and thus local VEP responses from areas within the visual field could be extracted from the EEG recorded in the occipital cortex. Thus the multifocal stimulus provides the ability to topographically analyse VEP recordings. The original visual stimulus used by Baseler resembled a series of hexagons with checkerboards

comprising of 24 equal triangles forming each hexagon. The hexagonal shapes were scaled such that the area of each hexagon increases in size with eccentricity proportional to cortical magnification of VEP signals. More commonly however a square dartboard pattern was used [99]. The pattern was controlled by a pseudo-random binary exchange at each of the test sites in the visual field. Thus the individual VEP signals may be extracted from the EEG by cross-correlation of the response evoked and the stimulus sequence. This system has since been modified and released commercially as the VERIS-Scientific™ system (Electro-Diagnostic Imaging Inc., San Francisco) [100].

Graham et al first examined the use of mVEP in glaucoma patients using a bipolar occipital straddle electrode positions. 42 glaucoma patients with reproducible visual field defects were tested. The mVEP showed loss of signal amplitude in the areas corresponding to the scotomas seen on Humphrey visual field analysis [101]. However, the amplitude difference between individuals was noted to be significant, thus limiting diagnostic ability of the mVEP. Using inter-eye asymmetry analysis to identify early defects, Graham et al. found that by comparing the test points between eyes for an individual 98.6% of patients with glaucomatous field loss could be identified. They also found that defects could be identified in some high risk glaucoma suspects in whom subjective perimetry was still normal [102].

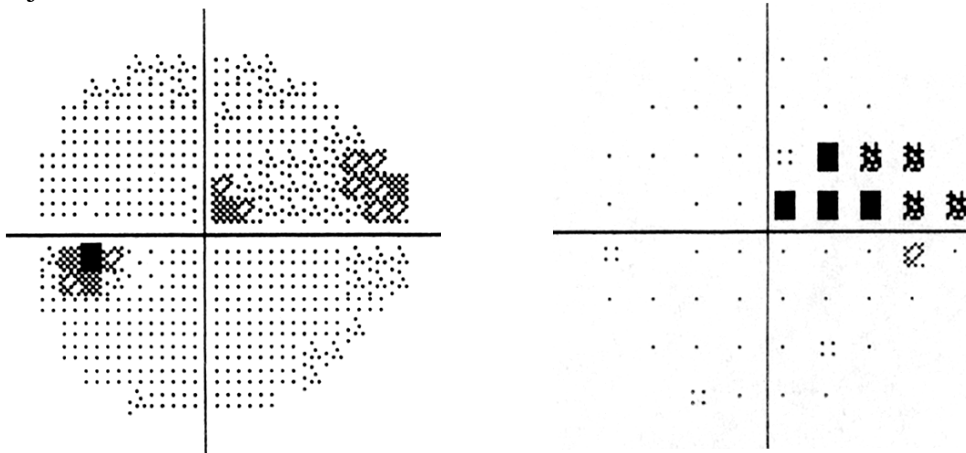
A second commercially available system, AccuMap™ (ObjectiVision Pty Ltd, Sydney) has been developed in Australia. This system uses 58 segments in a dartboard configuration, each containing 16 scaled square checks. The segments are cortically scaled with eccentricity to stimulate approximately equal areas of cortical (striate) surface. The system employs a spread spectrum technique using the Kassami families of binary sequences [101]. In a standard recording run of 54 seconds, each of the 58 segments of the visual field is stimulated approximately 2000 times. This technique also permits computation of the resulting signal by cross-correlation of the response evoked by the sequence stimulation, with the sequence itself at each of the stimulation sites [103]. There are some key differences between this system and the original VERIS™ system. Firstly, real time representation of the EEG signal allows the quality of the

recording to be checked during the acquisition of data. Secondly, the software has an inbuilt method for scaling the raw mVEP data according to the patient's background EEG and thus their own unique conduction characteristics. This reduced the problem of inter-subject amplitude variability [104]. The new electrode positions used, in the form of a cross around theinion, allows for recording across multiple channels, improving signal strength as the different recording vectors overcomes the problems of cortical convolution obstructing previous monopolar or single channel recordings [105]. Finally, a fixation target was added to the stimulus pattern to maintain patient attention and ensure central fixation [106].

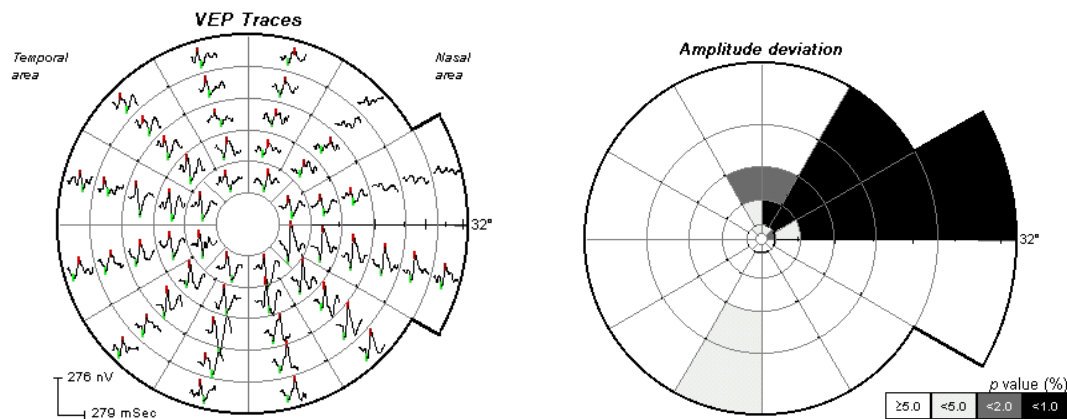
It has been established that this new multifocal VEP technique can reliably map visual field loss and that the findings strongly correlate with subjective perimetry results (see Figure 1.5) [102-104, 107-112].

Figure 1.5.

a) Subjective visual field, HVF, white-on-white.



b) Objective visual field, mfVEP – white on white.



The field of multifocal VEP glaucoma diagnosis has followed the amplitude changes seen in this optic neuropathy. This leaves us open to examine what other useful information may be gained by exploring latency analysis in different optic neuropathies.

ii. Why mVEP is more than just an objective perimeter

By allowing assessment of 58 domains of the visual cortex, the mVEP signal reflects the function of the visual pathway. To restrict the use of the mVEP to just one of “objective perimeter” does not make use of all the functionality it offers. The visual evoked potential measures both the signal amplitude and the time component of that signal, or latency. The mVEP is more than just a perimeter as it can detect evidence of disease long

after the recovery of the visual field as determined by amplitude analysis. In detecting latency delays along the visual pathways, the mVEP can map the “watermark” left by previous disease.

Glaucoma studies have examined the role of latency in glaucoma diagnosis and found 4% abnormality in the normal controls and only 10% abnormality in glaucoma patients [113]. Thus it would appear that the main role of latency analysis is in the diagnosis of inflammatory optic neuropathies, particularly when associated with Multiple Sclerosis.

The impetus for developing the mVEP for its applications to glaucoma came from the availability of treatment. Early intervention with medical glaucoma therapy reduces the progression of the disease [114]. With these figures the need for early, pre-clinical and objective detection became crucial. Thus the objective perimeter offered hope of achieving this level of diagnostic ability. To date there has been little interest in improving the pre-clinical diagnosis for a disease for which there is no treatment. This was the case in MS until the publication of the study by Jacobs et al[35] which showed that early intervention with interferon therapy reduced the risk of clinical progression from a clinically isolated syndrome, such as optic neuritis, to the full diagnosis of MS. Now there is an imperative to allow early diagnosis of those at the highest risk of MS, and be in a position to offer them this medication.

iii How mVEP may be used in optic neuritis and Multiple Sclerosis

Some of the limitations of conventional VEP recordings have been overcome with the advent of multifocal techniques in recording VEP signals [101, 105, 115-121]. Multifocal VEP (mVEP) provides a method to diagnose optic pathway conditions by assessing the VEP not as a single global response, but as responses from multiple individual segments of the field of vision. This allows for objective information on topographic visual field deficits (amplitude) to be combined with information on the speed of conduction along the visual pathways (latency).

The research on bifid VEP waves [87] suggests that this pattern is due to a splitting of the P100 into delayed and undelayed components. The multifocal technique will be able to separate out these two components based on spatial representation within the visual field. Patterns in this distribution of delayed and undelayed components may give further insight into the pathology and differences between demyelinating disease and other causes of ON.

The mVEP has been shown to be able to distinguish healthy volunteers from those with a history of demyelinating optic neuritis [122]. Hood et al used the multi-input procedure of Sutter to detect and track reduced and delayed mVEPs in patients with acute optic neuritis and multiple sclerosis [110, 123]. Using a different stimulus pattern paradigm in a multifocal VEP system, smaller amplitudes and latency delays were seen in MS patients with a history of optic neuritis, with a sensitivity of 92% and a false positive rate of 0% [124]. These early studies indicate that the mVEP could be of use in ON and MS, however further research is required.

Chapter 2

General Methods

A Subject recruitment

All patients were recruited from either Sydney Eye Hospital, St Vincent's Hospital or via contact with Multiple Sclerosis support groups. The control group was recruited from the general community as part of a larger project being undertaken at the Dept of Electrophysiology in the Save Sight Institute.

i. Optic neuritis group

- Acute: clinical diagnosis acute ON with unilateral visual loss, afferent pupillary defect and no other clinical cause or eye disease, within one month, confirmed by a consultant neuro-ophthalmologist
- Chronic/past: diagnosis of optic neuritis, confirmed by a consultant neuro-ophthalmologist at the time (unilateral visual loss, afferent papillary defect and pain on eye movement) more than one year previously
- Of those patients with MS: diagnosis confirmed by a consultant neurologist, fitting the McDonald classification of the disease, with MRI confirmation. Classification of MS type as either Relapsing Remitting (RR), Secondary Progressive (SP) or Primary Progressive (PP) forms of MS
- No other ocular pathology i.e. glaucoma, cataract, retinal detachment, other causes of optic atrophy (vascular, toxic, trauma, vitamin B12 deficiency)
- No other neurological pathology i.e. tumours, progressive neuropathies (Friedrich's ataxia, neurosyphilis)
- Age $15 < x < 60$
- Ability to sit and concentrate over period of approximately one hour
- No exclusions for visual acuity provided the patient can maintain focus on one point of a computer screen from 30cm. (i.e. ability to fixate in the central field $\sim 6/60$)

ii. Multiple sclerosis without ON

- Diagnosis of MS as confirmed by a consultant neurologist, fitting with the MacDonal classification.
- Patients were classified as either Relapsing Remitting (RR), Secondary Progressive (SP) or Primary Progressive (PP) forms of MS
- No recorded episode of optic neuritis.
- No history from the patient of an episode that could have represented optic neuritis before or after the diagnosis of MS.
- No other ocular pathology i.e. glaucoma, cataract, retinal detachment, other causes of optic atrophy (vascular, toxic, trauma, vitamin B12 deficiency)
- Age $15 < x < 60$
- Ability to sit and concentrate over period of approximately one hour
- No exclusions for visual acuity provided the patient can maintain focus on one point of a computer screen from 30cm. (i.e. ability to fixate in the central field ~6/60)

iii. Comparison group (ON as a differential diagnosis)

- Presentation to the emergency department with visual disturbance for which the attending physician included optic neuritis in the differential diagnosis
- Subsequent diagnosis of a condition other than optic neuritis, as determined by clinical history, disease course and other investigations
- Age $15 < x < 60$
- Ability to sit and concentrate over period of approximately one hour
- No exclusions for visual acuity provided the patient can maintain focus on one point of a computer screen from 30cm. (i.e. ability to fixate in the central field ~6/60)

iv. Control group

- Healthy normal individuals without ocular or neurological pathology as determined by a consultant ophthalmologist
- Perimetrically naïve

- Age matched
- Age $15 < x < 60$
- Ability to sit and concentrate over period of approximately one hour
- Normal corrected visual acuity (VAc 6/6)

Informed consent was obtained in all cases.

B Recordings

- Age
- Sex
- Date of episodes of optic neuritis, eye affected, and treatment received.
- Date of diagnosis of MS (if applicable), and current MS treatment (if any)
- McDonald criteria classification status
- Refraction
- Best corrected visual acuity*
- Colour vision, Ishihara*^
- Brightness perception – subjective, colour desaturation and brightness*
- Afferent pupillary defect*
- Optic disc appearance*
- Humphrey perimetry*^
- Accumap*^

When available

- MRI results – number and location of lesions

*Both eyes to be tested separately

^with appropriate refraction, and corrective additions for near vision

Though a review of the literature revealed much about the value of conventional VEP, the rigorous assessment of cVEP was not considered necessary for the purpose of this study. Our small sample size would add little to the current wealth of knowledge, and the focus of the study was to be mVEP which has been shown to have no direct correlation with cVEP. Limitations on access to cVEP recording devices also reduced the number of patients who had both cVEP and mVEP. A separate study analysing these two groups is currently underway.

C Visual assessment

i. General assessment

All patients underwent routine visual assessment. Best visual acuity was measured with refractive correction (if required) using a projected or wall mounted Snellen visual acuity chart and was recorded at 6 meters. Ishihara colour vision testing was performed using with 24 plate version of the test. Brightness perception testing was performed using the beam from a direct ophthalmoscope, and red desaturation was tested using a red test card. The patient was requested to score the perceived loss of brightness or redness in the affected eye as a percentage, where 100% was equivalent to the unaffected eye. The presence of a relative afferent pupillary defect (RAPD) was assessed using Levantin's swinging torch test. [125] Fundoscopy was performed with either direct ophthalmoscopy, slitlamp or binocular indirect ophthalmoscopy to look for optic disc swelling or pallor.

ii. Subjective perimetry

Subjective perimetry was performed using the SITA-FAST 24-2 protocol (Humphrey, San Leandro, CA).

iii Visual field analysis

Results from the Humphrey visual field testing were classified using the protocols described in the optic neuritis treatment trial.[12, 126] All tests had reliability indices within the normal limits (fixation loss <20%, false negative or positive errors <33%). The test was classified as abnormal in the affected eye if either the mean deviation or pattern standard deviation were beyond the normal limits ($P<0.05$) or if more than eight of the individual thresholds were beyond the normal limit ($P<0.05$).

D Multifocal visual evoked potentials

i. Test procedure

All mVEP testing was done with the Accumap™ system (Version 2.1) using standard stimulus conditions as described in previous papers [104, 112, 127-134]. All recordings were collected using monocular stimulation, with correction for near vision if required.

Four gold cup electrodes were placed in a cross position around theinion, allowing for 4 recording channels, representing the electrical vectors between the electrodes. In this way the effect of cortical convolution can be minimized [134].

The visual stimulus was generated on a computer screen (22" Hitachi high resolution display, stimulation rate 75 Hz). Fifty-eight closely packed segments in a dartboard configuration were used (see Figure 2.1). The segments were cortically scaled with eccentricity to stimulate approximately equal areas of cortical (striate) surface. Each segment contained a checkerboard pattern (16 checks) with the size of individual checks being proportional to the size of the segment and therefore also dependent on eccentricity.

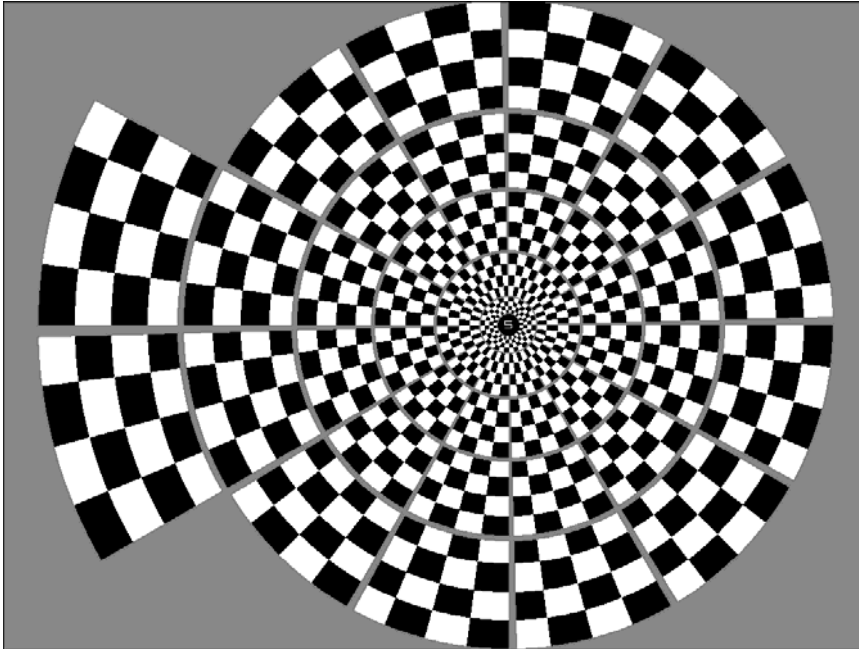


Figure 2.1: Visual stimulus checkerboard pattern, with central fixation target number displayed.

The system employed a spread spectrum technique using the Kassami families of binary sequences. Two opposite checkerboard pattern conditions underwent pseudo-random binary exchange at each of 58 sites in the visual field. Each input (stimulation site) was modulated in time according to a different binary sequence. In a standard recording run of 54 seconds, each of the 58 segments of the visual field was stimulated approximately 2000 times. The technique permits computation of the resulting signal by cross-correlation of the response evoked by the sequence stimulation, with the sequence itself at each of the stimulation sites.

Subjects were seated comfortably in a chair with the chin slightly elevated to relax neck muscles. They were asked to fixate on the small randomly changing number at the

center of the stimulus pattern. The distance to the screen was 30 cm, corresponding to a radius of the stimulus of 24° . Data was recorded using an ObjectiVision four-channel amplifier, with band-pass filter between 1 and 30Hz. The signal was amplified 100,000 times and then digitally filtered between 1 and 20Hz. The data-sampling rate was 450 Hz. Usually 7-9 runs of 55 sec each were recorded to provide a stable signal as indicated by the trace improvement software in the OPERA™ program.

For this study, standard stimulus conditions were luminance of the white check 146 cd/m^2 and luminance of the black check 1.1 cd/m^2 , producing Michelson contrast of 99%. Background luminance was maintained at a mean level of 73.5 cd/m^2 .

The Accumap™ system uses a fast Fourier analysis of all raw signals and after removing artefacts, ECG and alpha-rhythm patterns, scales the individual's VEP responses according to their background EEG levels [104]. This significantly reduces inter-individual variability seen with the VEP.

ii. mVEP analysis

Peak-to-trough amplitudes for each mVEP wave within the interval of 50-250 msec were determined and compared among channels for every stimulated segment of the visual field. The wave of maximal amplitude from each point in the field was automatically selected and the software created a combined topographic map (see Figure 2.2). The mVEP amplitudes in the combined trace array were compared with the normals database of 200 subjects to create a probability plot of possible scotomas. A scotoma was classified as an area with at least 3 adjacent abnormal points with at least one point having a deviation of $P < 0.1\%$, and the other 2 points $P < 0.5\%$.

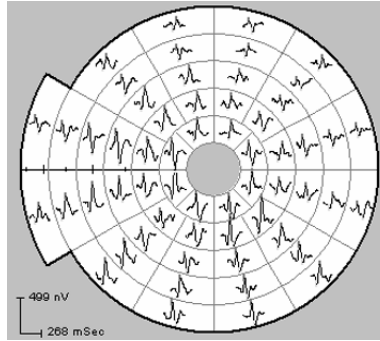


Figure 2.2: mVEP responses from normal subject corresponding to areas of visual field stimulated – right eye extending to 32 degrees eccentricity

The latency values were derived from the combined trace array, using Root Mean Square (RMS) values of the amplitude data to identify the timing of maximum power of the response in each trace and compared to normal database. A latency deviation plot can then be presented except at sites where the amplitudes are deemed too small for analysis (<60mV). A latency defect was defined as an area with 3 adjacent abnormal points with one point having a deviation of $P < 0.1\%$, and the other 2 points at least $P < 0.5\%$.

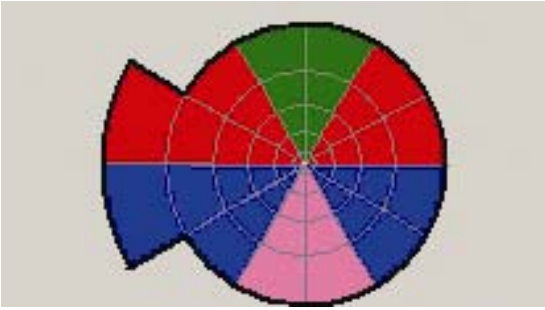
Inter-eye asymmetry analysis was also performed for both amplitude and latency values. Inter-eye latency asymmetry was included as an indicator of abnormality as this has been shown in several studies to improve the sensitivity of VEP [135-137]. The inter-eye asymmetry [102] was calculated for every segment of the tested visual field. A probability plot for asymmetry was constructed based on the normal database distribution of asymmetry between the two eyes. The definition of a scotoma for amplitude and latency asymmetry was similar to that used for the monocular analysis, described above.

Finally, the Accumap Severity Index (ASI) was calculated by the OPERA™ software for each subject. The ASI assigns scores to individual abnormal points and clusters of points with a weighting for location and whether they are present on the asymmetry plot (for more details see [112]). This score thus gives a guide for grading amplitude loss and is considered as being within normal limits for score between 0-11, borderline for score between score 11-19 and outside normal limit if score is 20 and higher.

Based on the definitions of abnormality for amplitude and latency scotomas, inter-eye asymmetry and ASI, described above, the mVEP tests were classified as either abnormal or normal.

Latency analysis was also performed on the average waveforms derived from the vertical channel for the four sectors of the visual field that produce mVEP signals of similar morphology [120]. As there is less variability over the population for the normal averaged sectoral latency values, it can show more subtle differences in latency for an individual when compared to the population. Figure 2.3 demonstrates these sectors (upper lateral, upper vertical, lower lateral and lower central) by colour coding. Based on trace waveform the maximum peak was measured for upper-lateral area and negative peak-for the remaining three areas. A z-score for latency deviation was derived for each of the four sectors. The z-scores for each sector were then averaged to give an overall latency deviation z-score for each eye of every subject. These were then compared between the two eyes. If the z-score asymmetry was outside normal limits (greater than 1.96 i.e. 5% probability) between the eyes, but the average latency z-scores for each eye were within normal limits (between -1.96 and $+1.96$ i.e. 95% probability), then the asymmetry z-score was used for the analysis.

a)



b)

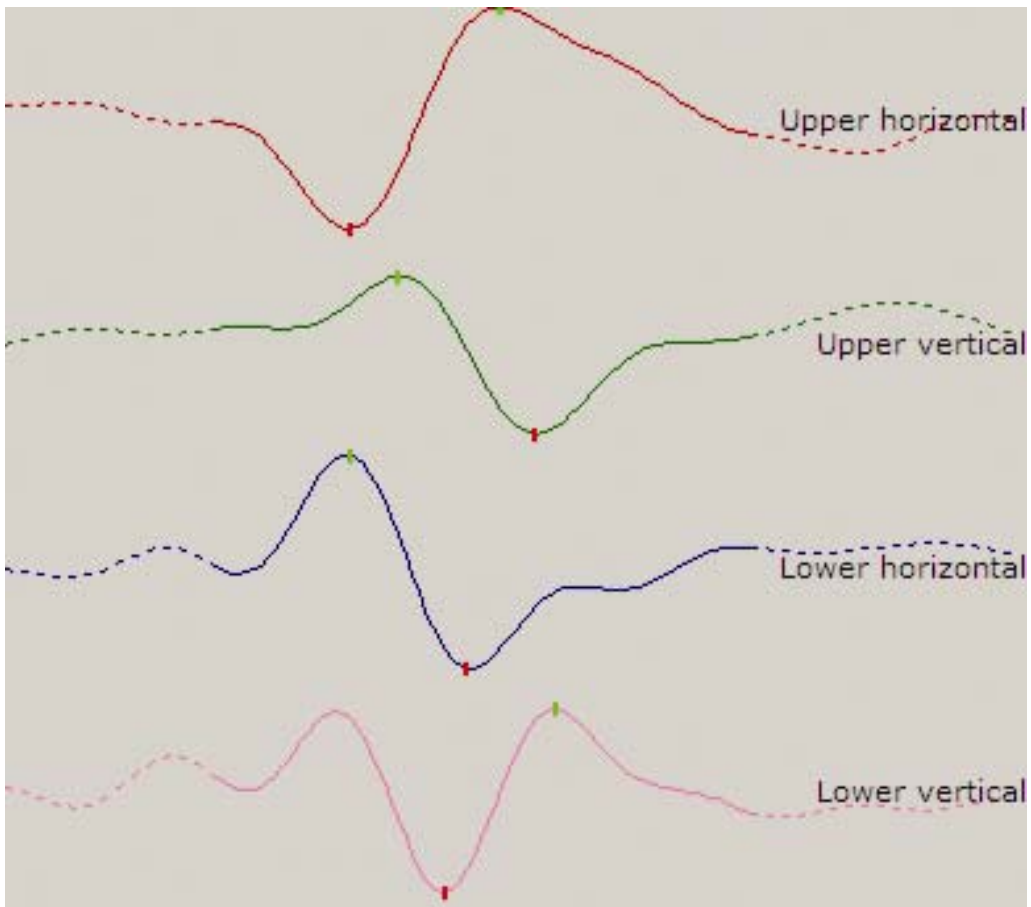


Figure 2.3:

a) graphic representation of the averaged sectoral waveforms.

b) The multifocal visual evoked potential field is divided into 4 sectors based on similar waveform morphology and colour coded.

E Statistical methods

All basic statistics were calculated using StatsDirect (StatsDirect Ltd, Cheshire, England).

In chapter 5 a multiple linear regression analysis was performed to ensure that no significant differences between the optic neuritis classification groups existed with regards to age, sex, time from diagnosis, treatment given and number of optic neuritis recurrences. ROC analysis was also performed with a latency z-score of ≤ 2 chosen as the optimal cut-off point between disease (MS) present and absent.

In chapter 6 the data was analysed using repeated measures ANOVA with the p-values adjusted for sphericity assumption using Huynh-Feldt correction.

Chapter 3

Sensitivity and specificity of mVEP in optic neuritis and comparison to other available tests

In this study, the results for the cross-sectional cohort are compared with normal controls, and patients with other causes for visual disturbance, in order to determine sensitivity and specificity of the mVEP in optic neuritis. The mVEP results are also compared to routine visual assessments in optic neuritis patients. This study showed that the mVEP was a viable method for assessing optic neuritis, as it had good sensitivity, specificity and corresponded with other indicators of visual dysfunction.

A Subjects examined

64 Subjects with inflammatory or demyelinating ON were recruited from the Sydney Eye Hospital and St Vincent's Hospital. The mean age of patients was 34.8 +/- 11.1 years. Each patient had a diagnosis of either acute or previous optic neuritis (with unilateral visual loss, afferent papillary defect and pain on eye movement) confirmed by a consultant neuro-ophthalmologist. The mean time from onset of optic neuritis was 16.6 months, with a range of 15 days - 5 years. Several patients had a history of past optic neuritis in both eyes (each eye on separate occasion) in which case each eye was included in the results. Patients with any other ocular pathology such as glaucoma, retinal abnormalities, ischemic optic neuropathy, toxic or metabolic causes of ON were excluded from this study. All acute ON patients were tested after completion of their IV steroid treatment.

A second group of 12 patients with MS but no recorded or subjective episodes of ON were recruited. The average age was 36.7 +/- 9.5 years. Of these patients 9 had a diagnosis of "relapsing-remitting" (RR) MS, 2 patients had "secondary progressive" (SP) MS and the last patient had a diagnosis of "primary progressive" (PP) MS.

Another group of 15 patients with a history of acute visual disturbance were recruited as a comparison group. In this group the average age was 38.2 +/- 10.3 years. These patients were referred from an emergency department with a differential diagnosis of ON as a cause for their visual disturbance. Subsequently each patient had ON ruled out based on clinical history, examination, other investigations and disease course. The most common subsequent diagnosis was “visual aura and migraine” in six patients. Two patients had a form of toxic optic neuropathy, one from toluene exposure and one from chemotherapeutic drugs. Three patients had other ocular causes for visual disturbance (infection, uveitis and increased intraocular pressure). Three patients had intracranial causes for visual disturbance, including idiopathic intracranial hypertension and 2 a final diagnosis of a transient ischemic attack. One patient was found to have an autoimmune optic neuropathy. No patient had a diagnosis of MS. All had normal cranial MRI scans. All were tested within the first 2 weeks of presentation to an emergency department. This group is referred to as the “differential diagnosis group”.

A control group of 20 normal patients was also tested, with their results compared to the built-in database. These patients were recruited from the general community and underwent examination by an ophthalmologist to exclude ocular disease. No patient had a diagnosis of MS. None of the patients had ever undergone perimetric testing prior to these tests. The average age was 38.0 +/- 7.6 years.

There was no statistically significant difference in ages between any of the 4 patient groups ($p > 0.5$, one way ANOVA)

B Ability to detect changes of optic neuritis

i. Amplitude

Deviation plots

The mVEP was able to detect amplitude deviation plot abnormalities in 90.7% of ON patients. These abnormalities consisted of amplitude reduction fulfilling the criteria of a scotoma in either the amplitude deviation or amplitude asymmetry plots (example: see Figure 3.1). The majority of the defects were classified as diffuse (53.1%), with central or centrocecal defects accounting for 21.9% and hemifield or altitudinal defects were found in 15.6%. There was no correlation between scotoma classification and time from diagnosis, ($p>0.9$, ANOVA).

Of those patients with MS, but no recorded episodes of ON, 25.0% (3/12) patients had severe amplitude deviation scotoma in both eyes. All patients with scotoma had a form of progressive MS, either primary or secondary progressive.

There were no amplitude deviation abnormalities detected in the control or differential diagnosis group.

Accumap Severity Index

When examining patients who had a definite episode of ON only 4 patients (6%) had a normal Accumap Severity Index (ASI) score. None of the control group had ASI abnormalities. Of the differential diagnosis group 4/15 (26%) had an ASI of greater than 20. However, all four of these patients had an ASI of <30 , indicating that the amplitude abnormality was minor, which is consistent with our finding that none had an amplitude deviation scotoma. In the MS group without ON, only the three patients with a progressive form of MS had ASI abnormalities.

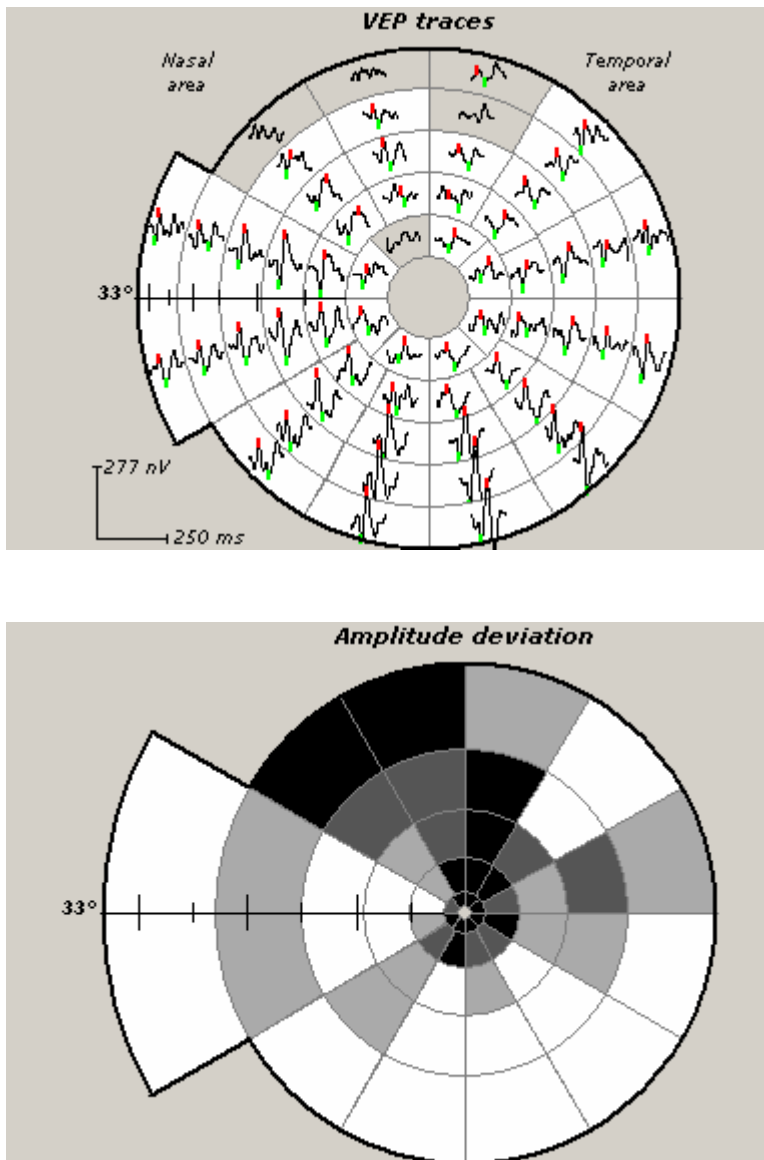


Figure 3.1: Acute optic neuritis amplitude loss seen on mVEP

- a) mVEP trace array, displaying all 58 VEP signals, smaller amplitude signals can be seen centrally and superiorly.
- b) Amplitude deviation plot, showing central and superior amplitude scotoma, with grey scale representing severity of amplitude loss in comparison to normal database.

ii. Latency

Of those patients with ON, 68.4% had a latency defect based on the definition of a scotoma, or sectoral latency z-scores (example: see Figure 3.2). Of the MS patients without ON 50% had a latency abnormality. Three of these patients had the relapsing remitting form of MS, the other three had either primary or secondary progressive MS.

None of the patients in the control or comparison group had a latency abnormality.

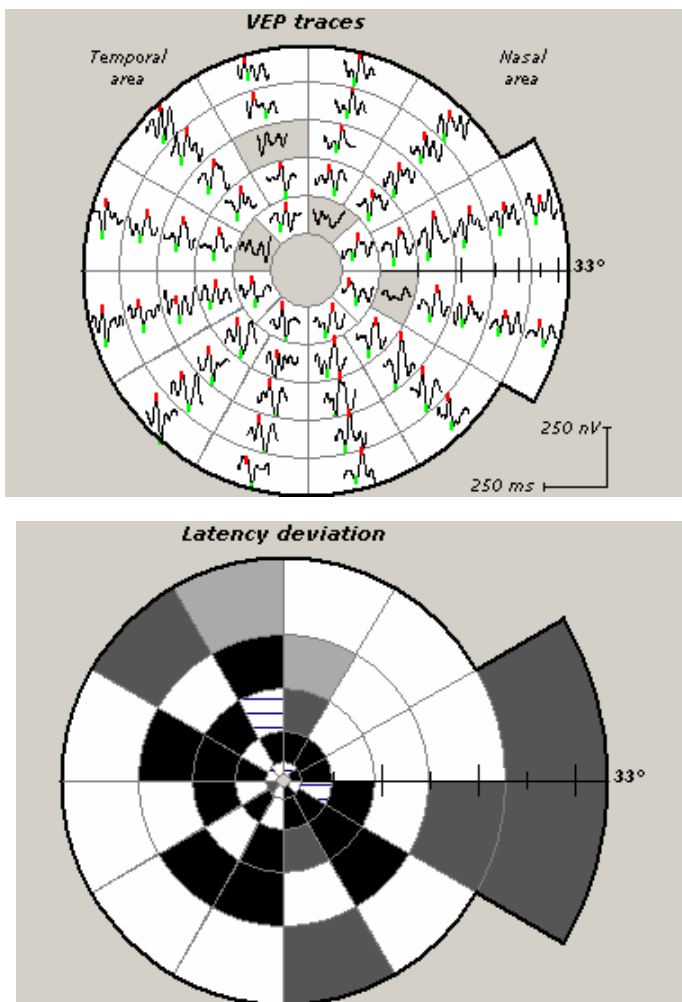


Figure 3.2: Acute optic neuritis latency delay

a) Trace array

b) Latency deviation plot, showing a latency scotoma throughout the visual field.

iii. Sensitivity and specificity

Amplitude

As the Accumap severity index (ASI) is a quantifiable measure of amplitude loss it was used in calculations of sensitivity and specificity. A cut-off score of greater than or equal to 20 was used to indicate abnormality. All patients with optic neuritis were compared with the combination of normal controls plus those in the differential diagnosis group and the MS patients without a history of optic neuritis. The sensitivity of ASI in detecting ON was 93.8%, with a specificity of 84.1% and a positive predictive value of 89.6%. A receiver-operator-characteristics (ROC) curve was calculated to determine the sensitivity and specificity of ASI in determining the presence of optic neuritis. The area under the ROC curve was 0.94 (figure 3.3)

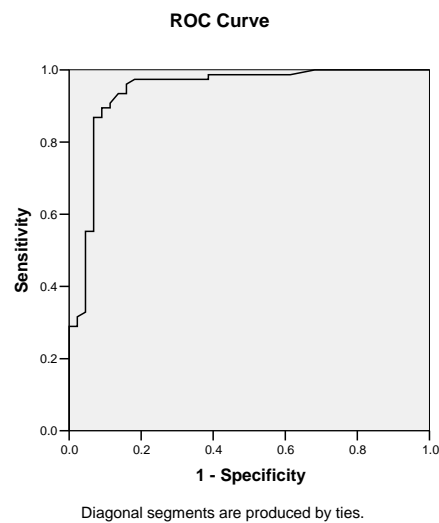


Figure 3.3: ROC curve of ASI in determining presence of optic neuritis.

Latency

The sensitivity and specificity of latency z-scores in determining the presence of optic neuritis was also calculated along with an ROC curve. The control and differential diagnosis groups' results were combined and compared to the ON patients. The MS patients without ON were not included, as other MS lesions along the optic pathways could cause prolonged latency results. The sensitivity of a latency z-score greater than or equal to 2 indicating the presence of optic neuritis was 65%, with a specificity of 100%, and a positive predictive value of 100%. The area under the ROC curve is 0.87 (figure 3.4).

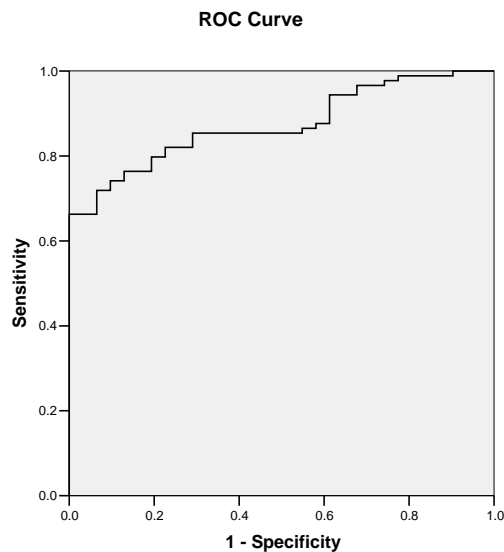


Figure 3.4: ROC curve of latency delay in determining the presence of optic neuritis.

C How does it compare with other available tests for ON patients

i. Visual acuity

Visual acuity (VA) was considered abnormal if vision was equal to or worse than 6/12. Abnormal visual acuity was seen in 42% of patients overall, in 50% of those within 6 months of their ON, and 35% of those beyond 6 months.

For those patients whose vision was equal to or worse than 6/12, the average ASI was 146, with a 95% confidence interval of 123-168. Of those patients with normal vision the mean ASI was 99, with a 95% confidence interval (CI 95%) of 145-119. Thus the ASI, which reflects overall amplitude abnormality, was significantly worse in those with a VA equal to or worse than 6/12, ($p=0.03$, ANOVA).

The mean latency z-score for patients with abnormal visual acuity was 2.3, (CI 95% 1.4 - 3.2). However, for patients with normal vision the mean z-score was 3.9, (CI 95% 3.1 - 4.6). Thus there was a statistically significant difference between the two groups ($p=0.007$, ANOVA). This may indicate that those with longer latency, and thus more likely to be the patients with MS, are the patients also most likely to recover vision fully due to the relapsing nature of the disease.

ii. Ishihara

An Ishihara colour vision 24 plate test score was classified as abnormal if the patient read less than 13 plates correctly on testing. Only 21% of patients had abnormal colour vision at the time of mVEP testing, with 16% abnormality within 6 months, and 25% of those beyond.

There was no correlation between the ASI or the latency z-scores and the ability to perform the colour vision test ($p=0.07$ and $p=0.29$ respectively, ANOVA). The correlation between ASI and colour vision neared significance at $p=0.07$. This is to be

expected as those with very poor vision, as reflected in ASI, will be less likely to be able to read the numbers on the Ishihara plates.

iii. Relative Afferent Pupillary Defect

A relative afferent papillary defect (RAPD) was classified as being either present or absent. The RAPD was not graded. A positive RAPD was detected in 50% of patients overall, 50% of those within 6 months of ON, and only 45% of those beyond.

There was a statistically significant difference found between the ASI scores of those with and without an RAPD ($p=0.001$, ANOVA). Of those patients who did have an RAPD, the mean ASI was 143, (CI 95% 126 – 134), whereas for those without an RAPD, the mean ASI was 94 (CI 95% 71 – 117).

There was no statistically significant difference in the latency z-scores between the two groups ($p=0.12$, ANOVA).

iv. Brightness perception

Abnormal brightness perception was classified as being present in any person who was able to notice a difference in the relative “brightness” of a pen torch. This test had the highest abnormality rate for all the ancillary visual tests of 54%, with 69% within 6 months of onset of ON, and 32% abnormal beyond 6 months.

There was a statistically significant difference found between the ASI scores of those with and without abnormal brightness perception ($p=0.016$, ANOVA). Of those patients who did have abnormal brightness perception, the mean ASI was 136, (CI 95% 117 – 154), whereas for those without brightness perception difficulties, the mean ASI was 98 (CI 95% 78 – 124).

There was no statistically significant difference in the latency z-scores between the two groups ($p=0.12$, ANOVA).

v. Red desaturation

Red desaturation was classified as being present in any person who could perceive a difference in the “redness” of the test card. Red desaturation was detected in 50% of patients overall, 50% of those within 6 months of ON, and only 45% of those beyond.

There was a statistically significant difference found between the ASI scores of those with and without an abnormal red desaturation test ($p=0.001$, ANOVA). Of those patients who did have red desaturation, the mean ASI was 142, (CI 95% 126 – 133), whereas for those without red desaturation, the mean ASI was 95 (CI 95% 72 – 117).

There was no statistically significant difference in the latency z-scores between the two groups ($p=0.12$, ANOVA).

vi. Humphrey Visual Field Analysis

An abnormal Humphrey visual field (HVF) was classified as per the ONTT (See Methods section). The HVF was abnormal in only 44.7% of patients tested overall, with 53% abnormal within 6 months of ON, and 37% abnormal beyond 6 months.

Those patients with an abnormal HVF were more likely to have higher ASI scores ($p=0.002$, ANOVA), with mean ASI of 145, (CI 95% 120 – 169). Those with normal HVF had an ASI of 96, (CI 95% 79 – 114).

There was no difference in the mean z-scores between those with normal and abnormal Humphrey visual fields ($p=0.12$, ANOVA).

D Discussion

i. Overall detection rates

The mVEP (amplitude or latency) was able to detect evidence of optic neuritis damage to the optic nerve in 96.1% of patients tested (90.7% amplitude, 68.4% latency). This corresponds well with the upper limit of reported amplitude sensitivity for full-field conventional VEP between 65-100% [14, 73, 75, 79, 85, 89, 90, 138]. Frequency of latency abnormalities in cVEP at follow-up of ON, ranges from 56 -100 % [79, 80, 85, 139]. The mVEP results from this study fall in the lower half of this range. Of the remaining 3.9% ON patients in whom no evidence of optic nerve damage was detected on mVEP, all had some abnormalities, but they did not reach the set criteria for a scotoma or latency delay used in this study.

A study in 2004 compared specificity of cVEP in detecting acute and chronic ON. Conventional VEP was found to be 70% sensitive and 12.5% specific in acute ON, compared to 75% sensitivity and 100% specificity in chronic ON [140]. Our study found an 84.1% specificity for mVEP ASI, and 100% specificity for latency delay.

Comparison between all VEP studies is difficult as each uses different entry and classification criteria. Secondly, VEP testing in each study was done at different times from the onset of the ON. This study aimed to give a cross-sectional view of all patients with optic neuritis past and present and the mVEP defects seen. This study proved that the mVEP is a viable test for detecting optic neuritis. Further analysis based on time from diagnosis will be performed in later chapters.

The amplitude deviation plots were sensitive in detecting visual field defects caused by ON, with no patients in the control or differential diagnosis group showing any scotoma. Only those MS patients with progressive forms of the disease who did not have a previous episode of ON had amplitude deviation scotomas. This probably represents ongoing axonal loss and atrophy causing the progressive nature of their disease.

The Accumap Severity Index (ASI) was the least sensitive amplitude measure for ON, as 26% of the patients in the differential diagnosis group, also had raised ASI scores. The ASI score is based on amplitude deviation and amplitude asymmetry, however individual deviated points that do not form part of a scotomatous cluster can cause this value to increase. Thus spurious readings from the peripheral field can result in a slightly abnormal ASI. In the 4 cases of abnormal ASI in the differential diagnosis group, all were <30 (abnormal being >20), indicating minor abnormalities.

All patients with a progressive form of MS showed multifocal VEP delay. This reflects the underlying pathology with a failure to recover full function after episodes of demyelination. Despite the lack of definite optic neuritis in these patients, the overall disease load in the cerebral cortex must at some point affect the optic radiations. It was surprising that more patients with relapsing-remitting MS did not show any evidence of latency delay. This would lead us to conclude that mVEP is specific though not sensitive for detected latency delay in those with MS who do not have a history of optic neuritis.

ii. Visual field defects seen on mVEP

The type of amplitude deviation scotoma seen on mVEP corresponds well to those reported using HVF analysis in the ONTT. We found 53.1% of patients had diffuse defects, 15.6% had altitudinal or hemifield defects, compared to 48% and 20% respectively in the ONTT. However we found that 21.9% of patients had an mVEP scotoma that was either central or centrocecal, compared to only 8% in the ONTT [126]. It must be noted however, that the figures from the ONTT come from patients with acute ON, whereas this study included patients months to years from diagnosis. It may be that the patients we are seeing now with ongoing central defects, had a more diffuse pattern closer to time of presentation, though we found no statistical correlation between scotoma type and time from diagnosis.

iii. Visual acuity

Poor visual acuity, classified as being equal to or worse than 6/12 was seen in 50% of patients within 6 months of their ON. In the ONTT vision worse than 6/9 was reported in 64% of acute patients. Most of our patients tested less than 6 months from diagnosis were tested within a month from diagnosis, thus these results seem consistent. However, we found 35% abnormality rates in visual acuity beyond 6 months, which does not fit with the reported rate of only 7% in the ONTT [13]. Given this is a cross-sectional study of patients visiting a tertiary hospital, it could be hypothesised that those most likely to return for follow-up beyond 6 months, are those with a worse visual outcome. This could represent a selection bias in our study.

Association has been seen between visual acuity and conventional VEP amplitudes in previous studies [17, 57, 141, 142]. These studies report that as visual acuity improved, the greater the whole-field or central cVEP amplitudes became. Our results on the mVEP confirmed this association as we found that ASI, which represents overall amplitude abnormality was significantly correlated with worse visual acuity ($p=0.03$, ANOVA). It is known that mVEP responses to the smaller central field checks are acuity dependant with an expected drop off at a visual acuity of 6/12 or worse, but the more peripheral zones with larger checks are not affected by acuity [143].

Reports on relation between latency values and visual acuity vary from no correlation, to longer latency values being associated with poorer visual function [17] and yet other studies reporting visual acuity is significantly better in cases with prolonged VEP latencies [144]. We report that the mVEP latency z-scores are higher, indicating prolonged latency, in those patients with normal vision, and that this is statistically different from the latency results seen in patients with decreased vision ($p=0.007$, ANOVA). It may be that as our study encompasses patients with past ON and MS, that these patients with longer latencies will have recovered better vision due to the remitting nature of the disease. Those patients with poor vision, may have developed optic atrophy secondary to an idiopathic ON, have a low probability of MS, and thus have normal latencies.

iv. Ishihara colour testing

The ONTT reported alterations in colour vision in 22% of patients at baseline [145], compared to 21% overall for our group (16% within 6 months, 25% after 6 months). We only performed the Ishihara colour plate test, which tests for red-green deficits. Thus we may have missed a proportion of patients with blue-yellow deficits. Despite this our results are comparable to other studies.

A study of 70 patients post ON showed that there was a 13% abnormality rate in the Ishihara colour test, and that this correlated to the degree of visual impairment, whereas it did not correlate to the VEP [14]. Other studies have reported that VEP amplitudes were associated with colour vision, and latency was not [141, 142]. In the early recovery phase of ON colour vision and VEP have been reported to be disturbed in a similar degree, whereas in the late recovery phase the VEPs remain more affected than colour vision [146]. We found no statistically significant correlation between the mVEP amplitude or latency values and the presence of colour vision deficits. However, the statistical association between ASI and colour vision, neared significance at $p=0.07$ (ANOVA). It is possible that with the inclusion of blue-yellow testing, a correlation may have been seen.

v. Relative afferent papillary defect

The levels of positive RAPD seen in this study, 50% of patients, were low in comparison to other studies. In 1981 Cox et al reported a 92% rate of positive RAPD in patients with recovered unilateral optic neuritis [147]. They further examined the relationship of the relative afferent papillary defect and the VEP. They found a correlation between the RAPD and VEP amplitude in patients with anterior ischemic optic neuropathy but not in patients with optic neuritis. There was however a weak correlation between pupillary response and VEP latency [125]. A group in Japan, looking at patients recovering from ON, however found that the degree of RAPD significantly correlated with a reduction in VEP and visual acuity, but did not correlate with delay of VEP latency [39]. Our study

found a statistically significant correlation between decreased mVEP amplitudes and the presence of an RAPD, no such correlation was found with latency. This is consistent with the physiology of the afferent papillary defect, in that smaller amplitudes represent a relative scotoma in the affected eye, thus producing the RAPD effect.

vi. Brightness perception

Brightness perception abnormalities were only seen in 54% of patients overall (69% within 6 months of diagnosis, and 32% beyond 6 months). Other studies however have reported up to 89% abnormality rates in brightness perception at more than 6 months from the time of diagnosis of ON [42].

Consistent with the findings for RAPD we found that abnormal brightness perception was significantly correlated with decreased mVEP amplitudes, but not delayed latency.

Searching papers with Medline search terms “VEP” and “optic neuritis”, no reports were found correlating cVEP results with the brightness perception test used in this study. One study did find that cVEP latency delay correlated with a delayed visual perception of altered light intensity [148].

vii. Red desaturation

Red desaturation was seen in 50% of patients overall (50% of those within 6 months of ON, and only 45% of those beyond).

Similar to the findings for RAPD and brightness perception, we found that the presence of red desaturation was significantly correlated with loss of mVEP amplitudes, but not latency delay. There were no journal articles found using Medline that discussed the presence of red desaturation in optic neuritis, and the relation to VEP findings.

viii. Humphrey visual field analysis

Though exact classification of HVF defect types as per the ONTT protocols was beyond the scope of this study, ONTT guidelines were used to classify the HVF as either normal or abnormal. Thus correlation between the mVEP visual field amplitude scotoma and the HVF scotoma type was not performed.

The entry criteria for the ONTT included an abnormal HVF, and our study did not. Thus all HVF abnormality results should be proportionally lower. Overall abnormality rates were 44.7% (53% within 6 months, and 37% beyond 6 months). At the six month mark in the ONTT 49% of HVF were abnormal, with 44.1% abnormal at one year. Thus given that the average time from diagnosis in this study was 16.6 months, the overall abnormality rates are within the range that could be extrapolated from the ONTT results.

Exact point by point comparison of mVEP amplitudes to HVF test locations cannot be performed as the test locations do not fall in the same place. However it has been noted that regions of depressed responses to the HVF in patients with ON correspond to substantially smaller mVEP amplitudes. In the same study Hood et al noted that post ON, delayed responses were seen in areas of previous visual field loss. However, no relation between latency and HVF was made[110]. Our results confirm that a loss of amplitude or increased ASI is correlated with abnormality on HVF testing ($p=0.002$, ANOVA). There was no relationship found with mVEP latency and HVF results.

ix. Conclusion

In this series of optic neuritis patients the mVEP amplitudes correlated with visual acuity, relative afferent papillary defect, brightness perception, red desaturation and HVF abnormalities. Multifocal VEP latency delay was only associated with improved visual acuity.

Overall the mVEP is both sensitive and specific in detecting damage due to optic neuritis, past or present. It is thus reasonable to investigate the further use of the mVEP in patients with optic neuritis. These results convey only the overall comparison of the mVEP performance with various other visual acuity tests, more detailed analysis may reveal further associations.

Chapter 4

Reproducibility of multifocal VEP

Once it was established that the mVEP could reliably detect the changes of optic neuritis, it was necessary to calculate whether the test-retest variability made it a viable method for following patients over time.

Amplitude reproducibility data has already been studied in the mVEP. It has been reported that for a change in amplitude to be reliably identified, it would have to alter by 30-40% [149]. However, with scaling and cluster analysis the point-by-point amplitude variability in the normal population can be reduced to 20%. The difference between the cut-off points (<5%, <2% and <1%) for significant variation from population means, as shown on the amplitude deviation plot are 1.95SD, 2.5SD and 3.4SD, equivalent to a 15% change in amplitude. Thus, if the normal variation for the population on repeat testing is allowed, then this normal variation could alter the reported severity of a patient's amplitude defect. Wall and colleagues have also reported poor mVEP amplitude reproducibility at the margins of glaucomatous scotomas in patients with moderate to severe visual loss [150]. For scotomas of increasing density, the reproducibility results worsen.

Other factors that can contribute to poor reproducibility could include fixation shifts. It has been shown that a 1 degree fixation error can produce a large decrease in the signal amplitude at the foveal region of up to 60%. The size of this effect drops rapidly beyond 3 degrees of eccentricity [151]. This can be a particular problem in optic neuritis patients who frequently have a central scotoma. Secondly, optimal refraction must be obtained, as the central amplitude recordings are acuity dependant. When defocus is at 2.0+ dioptres, the central mVEP amplitude responses are reduced by approximately 60%, however beyond 7 degrees of eccentricity there is no effect of the defocus [152]. There are obvious technical problems that can cause and decrease in the signal to noise ratio and

thus reduce the reliability and reproducibility. These include poor electrode positioning and skin contact, excess noise from muscular activity in the neck and excess alpha rhythm. With adequate technician training, these last few problems can be easily avoided. However, a gaze fixation device would be required before fixation shifts could be monitored. The significance of all these problems becomes apparent when attempting to perform follow-up studies for patients with known visual field defects.

It has been noted that the retest variability of visual field defects for patients with optic neuritis is very high when using the Humphrey Visual Field analyser. Wall et al. tested optic neuritis patients 5 times within one day, finding that visual field defects ranged from hemianopia to normal to central scotomas for an individual patient. It was concluded that this variability was an intrinsic problem with the HVF algorithms, rather than a real variability in the patient's pathology [153]. The step-wise incremental changes in target luminance are dependant on the subjects initial responses in the four cardinal points. Thus error in one of these four points can cause significant alteration to the rest of the results.

Firstly, reproducibility of latency results in normals needed to be determined to supplement the knowledge of amplitude reproducibility. Secondly, given this known problem with perimetric methods in patients with optic neuritis, a simple reproducibility test was performed on optic neuritis patients using the mVEP looking at both amplitude and latency..

A Latency reproducibility in normal patients

i. Methods

20 patients were recruited from the general community for repeat mVEP testing in order to determine the test reproducibility. To ensure that there was no subclinical ophthalmic or neurological pathology a consultant ophthalmologist reviewed the patients. The

patients were age matched to those recruited for the optic neuritis study. There was a female to male ratio of 6:1, with an average age of 36.8 +/- 7.7. There was no statistically significant difference between the ages of this group and the study population ($p > 0.1$, Student's T-test).

The average time between tests was 1.5 months +/- 0.6 months.

Latency data was taken from the mVEP trace arrays across all 58 points in the visual field for each patient. Although both eyes were tested the right eye only was used for analysis. A Coefficient of Variability (CV) was calculated for each location as follows:

$$CV = ((\text{latency1} - \text{latency2}) / \text{latency1}) * 100$$

Standard deviation of CV's was calculated to assess the variability for each area of the visual field. The results were then averaged by rings based on eccentricity from the central fixation point. The rings were labelled from the central fixation point, see Figure 4.1.

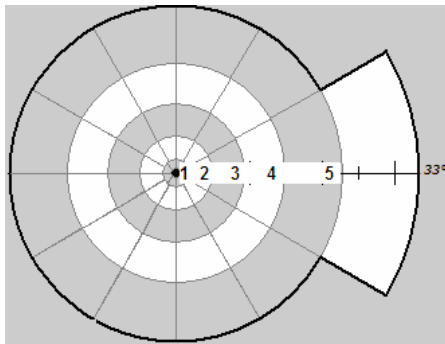


Figure 4.1: Numbering system used for labelling multifocal VEP trace array rings.

Latency data was also taken from the sectoral waveforms. The sectoral averaging occurs across points with similar waveform morphology (See Methods). The latency of the wave peak was taken for the upper horizontal sector (UH), and the latency of the wave trough was taken for the other 3 sectors (upper central – UC, lower horizontal – LH, lower central LC). This usually corresponds to the second deflection point of the mVEP

wave. A Coefficient of Variability was calculated in a similar manner to that shown above.

ii. Latency deviation

There was no statistically significant difference between the latencies values from each segment in the visual field from the first test to the second test, ($p=0.41$, paired T-test).

The latency values for each of the 58 points in the visual field were compared for each patient between two tests one month apart. The average percentage change for latency values was $13.7 \pm 58\%$. With an average latency value of 125.8ms, this is equivalent to a 17 ± 72.5 ms difference between points.

Accurate latency readings are dependant on signal amplitudes of sufficient strength. It is known that signal amplitude decreases in the outer rings of the mVEP [149]. Thus to investigate if increase in variability with eccentricity is due to signal strength, latency variability was plotted against the amplitude for each point tested in the visual field. There is a significant association between amplitude and latency variability ($t_{630}=-8.5$, $p<0.001$, linear regression). For every increase in amplitude by 10mV, latency variability decreases by 0.5%. See Figure 4.2.

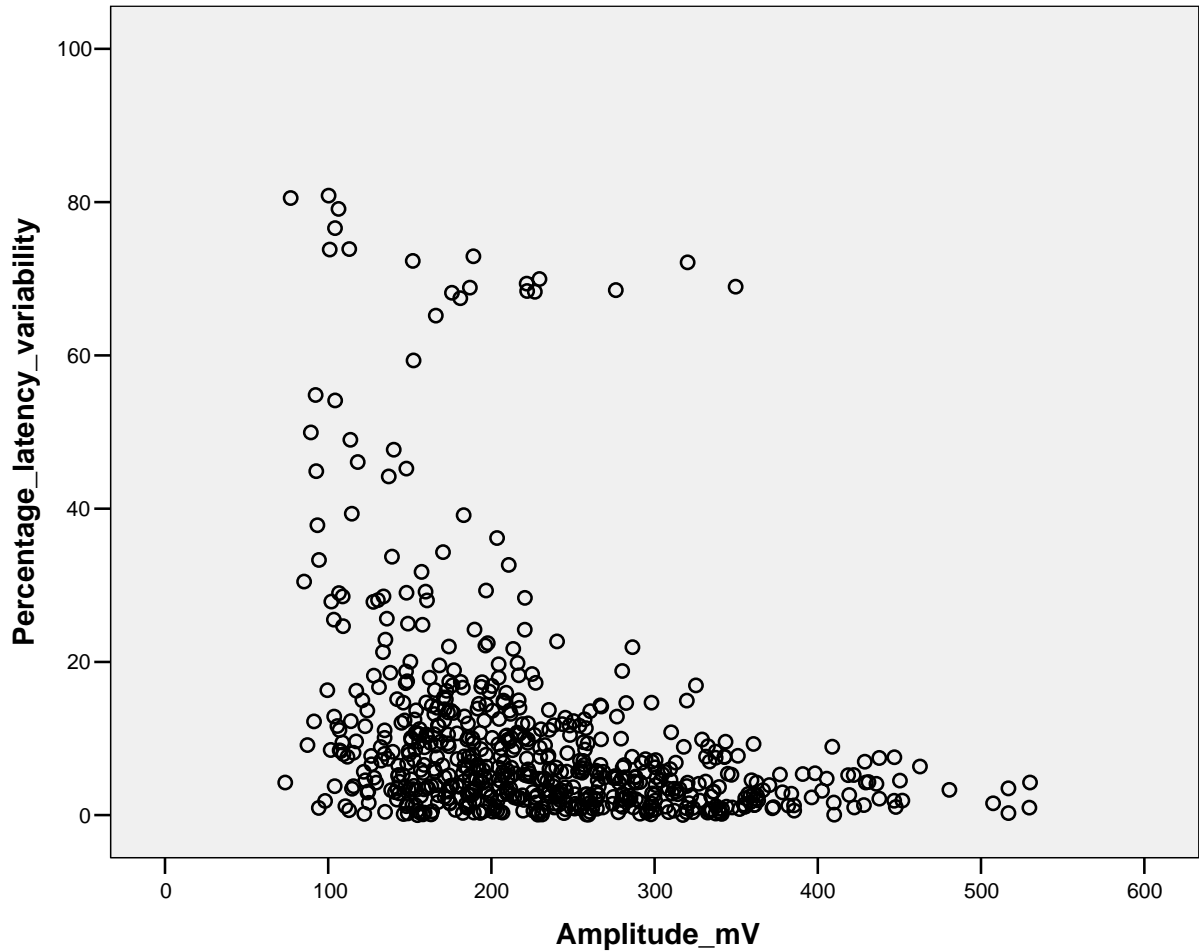


Figure 4.2: Variability of latency values as a percentage in relation to signal amplitude in millivolts (mV)

As amplitude variation increases with eccentricity from the fixation point, latency variability was averaged based on eccentricity. The latency variation point to point increased with eccentricity from the fixation point. The variation for the points in the central ring was only $5.4 \pm 1.6\%$ over an average of 123ms, or 6.5 ± 2 ms. The

variation in the outer ring was an average of 21.8 +/-22 %, or 27 +/- 28 ms. There was a steady increase in percentage variability between the inner and outer rings. See figure 4.3.

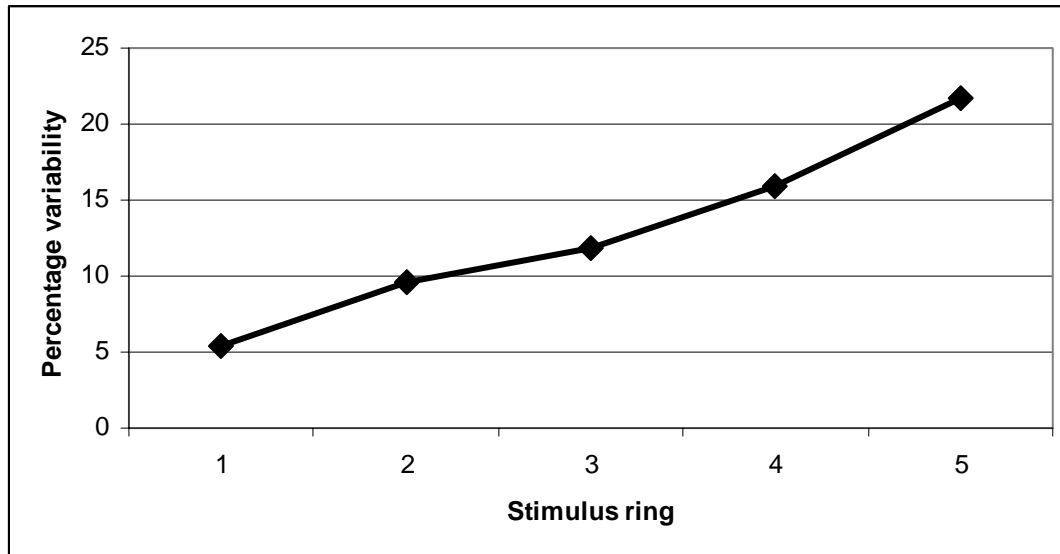


Figure 4.3: Increasing point by point latency reproducibility with eccentricity from the central fixation point.

iii. Sectoral analysis

There was no statistically significant difference between the latencies of any sector from the first test to the second, ($p > 0.5$, paired T-test).

The sectoral latency values for each patient were compared for each sector between the two tests for each eye separately.

The upper central and lower central sectors (UC, LC) showed the most variability between tests in each eye. The upper central field variation was on average $-0.6\% \pm 2.0\%$, with a range of $+7\text{ms}$ to -7ms . In the lower central field variation averaged $0.7\% \pm 2.7\%$, with a range of $+9\text{ms}$ to -9ms . However in the more reliable lateral sectors, had an average variation of $-0.06\% \pm 1.3\%$ and $0.2\% \pm 1.4\%$ for upper and lower horizontal (UH, LH) sectors respectively, with a range of only $+4\text{ms}$ to -4ms .

Thus the overall percentage variability for latency is 0.2% +/- 1.89%. Thus a patient whose latency varies by more than 4% (>2 standard deviations of variability) has shown a significant alteration in that latency. For each sector this equates to a change of 6ms, 7ms, 6ms and 5ms respectively for UH, UC, LH and LC sectors. In order to detect clinically significant changes in latency values, and to interpret consecutive test results, analysis of sectoral latency is more clinically viable than analysis made on point variability or eccentricity.

Due to the increased reliability and reproducibility of sectoral latency values, as well as the narrow normal population variation when compared to point latency values, further analysis of latency used in subsequent experiments focuses on this method.

B Test-retest reproducibility for patients with optic neuritis

i. Subjects

Five patients with known amplitude defects due to optic neuritis were asked to undergo repeat mVEP tests, one hour apart. Between tests the electrode headset was removed and the patients were given the opportunity to rest to reduce the risk of increased alpha rhythm during the second test. Patients were asked not to have a hot drink between tests, in order to avoid Uhthoff's phenomenon.

5 subjects participated with a mean age of 32.4 years. There were 3 females and 2 males. One patient has optic atrophy of 18 months duration following an episode of optic neuritis. Four patients have ON secondary to MS.

ii. Methods

Amplitude probability plots were examined for the eye with a known history of ON. The number of points with amplitude defects in the ranges of $p < 1\%$, $p < 2\%$ (includes $p < 2\%$) and $p < 5\%$ (includes $p < 2\%$, $p < 1\%$) were counted for each test. Each point was given a weighted value based on deviation from the inbuilt data base, with points of amplitude defects $p < 1\%$ scoring highest. This served as a surrogate ASI score. As amplitude asymmetry was not included in this study ASI could not be used. The difference between this surrogate amplitude scores between the two tests for each eye was also examined. The number of points which were abnormal on the repeat test counted for each probability level. A ratio, expressed as a percentage, for the number of points missed versus the number of points in common was calculated. This gives an indication of the overall pattern accuracy of the test.

Sectoral latency results were also examined to determine the Coefficient of Variance (CV) seen between tests.

$$CV = ((\text{latency1} - \text{latency2}) / \text{latency1}) * 100$$

iii. Amplitude deviation

The overall amplitude deficit pattern was similar on each test. 3 patients showed central scotomas, one more predominantly altitudinal defect and the other more temporal – these pattern classifications were reproducible.

The average difference between the amplitude scores of the different tests was 4.3% with an overall 2 standard deviation variance of 8.9%. The surrogate amplitude score used in this study is comparable to the ASI calculated automatically by the system. Therefore the normal fluctuation to be expected is less than 8.9%. Thus between tests, a clinician would have to see an alteration of ASI >9% in order to diagnose a significant alteration in amplitudes.

When examining the topography of the points with decreased amplitudes as seen on the amplitude deviation plot, we calculated the percentage of points which the two tests had in common. At the significance levels of $p < 1\%$ (black squares), $p < 2\%$ (dark grey squares) and $p < 5\%$ (pale grey squares), there was a 49 +/- 19%, 75 +/- 18% and 82 +/- 10% concordance between the two tests respectively. Therefore the overall shape of the scotoma would remain more or less the same, whereas the depth or level of abnormality was less reliable.

iv. Sectoral latency

There was an average difference of 4.1 +/- 2.2ms between the sectoral latency values between tests for each patient. These results equate to an average coefficient of variance of 2.3 +/- 2.9%.

For each of the sectors, upper horizontal, upper vertical, lower horizontal and lower vertical there was a respective difference in latency z-scores of 1.6%, 1%, 4.7% and 0.5% respectively.

C Discussion

In order to assess if this technology, which has been shown to accurately detect the presence of optic neuritis, is of real clinical use, it is important to know the level of variability created by the method of testing. This allows clinicians to use the test on a repeat basis to follow the course of disease and adjust management as appropriate. The amplitude variability has been studied in normal patients [149]. Thus the latency reproducibility in normal patients was assessed, firstly based on the eccentricity of the points from the central fixation point, and then by sectoral averaging.

By comparing the individual test points on the mVEP between one test and another, it was found that results varied by a standard deviation of +/-58% (or 69msec). When taking into account that to be of clinical significance, a test result would have to change by more than 2 standard deviations, this results in a requirement for changes of more than 146ms. This does not make practical sense in a clinical setting. It has been shown that latency variability decreases with mVEP signals of greater amplitude. Thus in order to improve the level of variability, methods to improve the signal to noise ratio are required.

When examining the variability of sectoral latencies, it is apparent that this method of assessing latency is more clinically practical. Sectoral latencies were more stable than the point latencies, with a variability of 0.2 +/- 1.9%. Thus in order to diagnose an alteration in latency, based on a variation of more than 2 standard deviations, a clinician would only have to look for changes of more than 6ms in the upper horizontal and lower horizontal sectors, and 7ms or 5ms in the upper and lower central sectors respectively. These values compare favourably with reports of conventional VEP latency variability,

which state that the P100 must alter by more than 6 – 12ms to be clinically significant [154-156].

When examining patients with a visual field defect secondary to optic neuritis, we found less variability than has been reported for ON patients based on the HVF. However, we only tested patients twice in one day, compared to the 5 times used by Wall et al [153]. As the ASI remains normal in the normal control patients, the variability of ASI was determined based on the optic neuritis patients. It was found that a variation of ASI by more than 9% would be required for a clinician to be certain of an overall change in the amplitude deviation of the mVEP. However, this was based on patients with an average ASI of 180, which is significantly abnormal (normal being <20). Thus it is not certain whether this value of 9% could be extrapolated to patients with ASI values closer to the normal range.

The overall shape of the visual field defect was reproducible, however the depth or severity of the deficit was not. There was an 82 +/-10% concordance between the two tests, for any particular point to show some degree of amplitude deviation (i.e. points with $p < 5\%$). However for points of greatest deviation, $p < 1\%$, there was only 49 +/-19% agreement between tests. In other words if a point on an amplitude deviation plot was coloured black (meaning $p < 1\%$) in one test, there was only about a 50% chance, it would be coloured black in a test done only one to two hours later. However there is an 82% chance that it would still have an amplitude deviation of at least $p < 5\%$, or shaded pale grey. Thus a clinician could only really comment on changes in the overall amplitude deviation pattern between tests, not the severity of the defect. An improvement or worsening of severity would have to be present on more than one repeat test in order to approach clinical significance.

Sectoral latency reproducibility was the same as seen for normal control patients. Thus any alteration of sectoral latency by more than 5-7ms (depending on the sector) could be classified as clinically significant.

Thus the multifocal VEP can be used to track overall amplitude deviation changes and sectoral latency changes between tests in patients with optic neuritis.

Chapter 5

Correlation of Multiple Sclerosis risk assessment with mVEP findings

(Sections A-D: Published as “Multifocal visual evoked potential analysis of inflammatory or demyelinating optic neuritis”. Fraser, C; Klistorner, A; Graham, S; Garrick, R; Billson, F; Grigg, J. Ophthalmology. February, 2006 in press)

Objective: To determine the sensitivity of multifocal visual evoked potentials (mVEP) in optic neuritis of an inflammatory or demyelinating nature.

Design: Cross-sectional study.

Participants: 64 patients with a confirmed diagnosis of optic neuritis (past and acute) participated in the study. Based on the McDonald Multiple Sclerosis (MS) criteria 25 patients (27 eyes with ON) were deemed to have isolated optic neuritis and thus not have MS (“not-MS” group), and 19 patients (24 eyes with ON) had a diagnosis of MS (“MS” group) at the time of testing. The remaining 20 patients (25 eyes with ON) were at a high risk of MS, but diagnostic evaluation was equivocal, and thus they were classified as the “possible MS” group. A control group of 20 normal patients was also enrolled.

Testing: Multifocal VEP was performed. All ON patients had recent magnetic resonance imaging scan (MRI) imaging of the brain and spinal cord.

Main outcome measure: Multifocal VEP amplitude and latency values were analysed within each group and compared to the normal controls.

Results: No abnormality was recorded on mVEP in the control group. Of all the ON eyes, 74 (97.3%) were abnormal on mVEP testing. Amplitude values were abnormal in 92.6% of “not-MS” eyes, 92.0% of “possible MS” and 100% of those with “MS”.

Latency was abnormal in 33.3%, 76.0% and 100% respectively. There was a significant difference in the mVEP latency z-scores among all ON groups ($p < 0.01$, Kruskal-Wallis). While distribution graphs of latency z-scores in “not-MS” and “MS” groups had single peaks and were clearly separate from each other, the latency z-score distribution within

the “possible MS group” in post-acute patients was bimodal with each peak corresponding to the distribution of the “not-MS” and “MS group” respectively. The mVEP latency z-scores had a sensitivity and specificity of 100% in detecting patients with ON due to MS when compared normal patients.

Conclusion: The mVEP is a sensitive and specific tool for detecting optic neuritis. There was a significant difference in latency analysis findings between patient groups classified according to the McDonald MS criteria.

Significance: Latency results suggest a role in identifying a patient’s risk for future MS.

A Subjects

64 Subjects (mean age 34.8 +/- 11.1 years) with inflammatory or demyelinating ON were recruited from the Sydney Eye Hospital and St Vincent’s Hospital. Each patient had a diagnosis of either acute or previous optic neuritis (with unilateral visual loss, afferent papillary defect and pain on eye movement) confirmed by a consultant neuro-ophthalmologist. The time from onset of optic neuritis was recorded (mean 16.6 months, range 15 days - 5 years). Several patients had a history of past optic neuritis in both eyes (each eye on separate occasion). Patients with any other ocular pathology such as glaucoma, retinal abnormalities, ischemic optic neuropathy, toxic or metabolic causes of ON were excluded from this study.

Those patients who received pulse IV methylprednisolone were documented. All acute ON patients were tested after completion of their IV steroid treatment.

Magnetic resonance imaging scans had been performed on all patients. Based on further clinical history and examination, as well as cranial and spinal cord MRI, the ON patients were classified as either “not-MS”, “possible MS” and definite multiple sclerosis (“MS”) using the McDonald criteria [30]. “Not-MS” patients were those with a normal MRI and evidence of an inflammatory cause for their ON (for example: history of viral illness, elevated lymphocyte count, isolated optic nerve enhancement on MRI). Though these

patients are called “not-MS” by the McDonald criteria, this is a reflection of their clinical condition at the time of testing and does not indicate that these patients will never develop MS in the future [157]. “Possible MS” patients had an attack of optic neuritis, some white matter lesions seen on MRI (not fulfilling the Barkhof criteria for MS), but no other neurological symptoms, signs or previous episodes that were sufficient to fulfil the criteria for definite MS. All “MS” patients fulfil the McDonald criteria for definite multiple sclerosis.

There were 25 patients with ON (27 eyes) classified as “not MS” (or low risk for future MS [30]) with a 7:2 ratio of females to males and an average age of 35.6+/-9.6 years. The average time from diagnosis of optic neuritis was 13 months (range 15 days – 5 years).

In the “possible MS” group of 20 patients (25 eyes), the average age was 34.8+/-11.9 years, with a female to male ratio of 3:1, and an average time from diagnosis of 13 months (range 15 days – 4 years).

The 19 ON patients (24 eyes) who were classified in the MS group had an average age of 36.0 +/- 11.8 years, a female to male ratio of 3:2 and an average time from diagnosis of 28 months (range 15 days - 4 years).

There was no statistically significant difference between the ages of the patient groups, ($p>0.5$, ANOVA).

A control group of 20 normal patients was also tested, with their results compared to the built-in database. These patients were recruited from the general community and underwent examination by an ophthalmologist to exclude ocular disease. No patient had a diagnosis of MS. None of the patients had ever undergone perimetric testing prior to these tests. The average age was 38.0 +/- 7.6 years. There was no statistically significant difference in ages between any of the 4 patient groups ($p>0.5$, ANOVA).

B Amplitude findings

Table 5.1 shows the number of ON eyes with an amplitude abnormality on mVEP. As can be seen from the table, the percentage of eyes with an amplitude deviation scotoma increased over “not-MS” to “possible MS” and “MS” groups. There were no amplitude abnormalities seen in the control group.

	Not-MS	Possible MS	MS
Amplitude deviation	70.4%	68.0%	91.2%
Amplitude asymmetry	85.2%	88.0%	79.2%
Abnormal ASI	92.6%	92.0%	100%

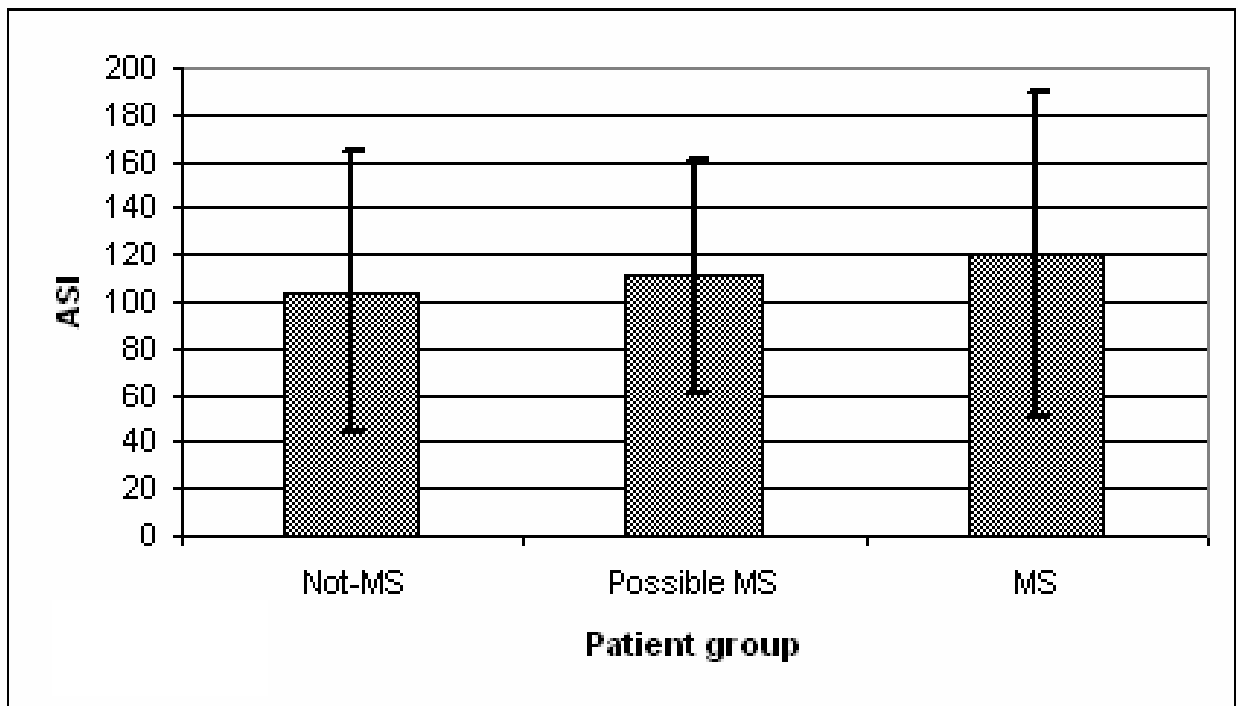
Table 5.1: The percentage of eyes within each patient classification group with abnormalities of amplitude deviation, amplitude asymmetry and an abnormal Accumap Severity Index (ASI).

Within the “not-MS” and “possible-MS” groups only approximately 70% of the eyes showed an amplitude deviation with respect to the normal population, however nearly 90% had inter-eye amplitude asymmetry, which indicates smaller but still detectable mVEP amplitude changes in a majority of eyes in these groups.

The rates of amplitude asymmetry decreased in patients with “MS”, which was probably due to an increased rate of bilateral disease, either clinical or sub-clinical.

The average ASI gradually increased from the “not-MS”, through the “possible MS” to the “MS” risk groups, this trend was not statistically significant ($p > 0.1$, one way ANOVA. (See Figure 5.1).

Figure 5.1: The average Accumap Severity Index (ASI) indicated the severity of amplitude loss. Though there is a trend for increasing ASI scores across the three patient groups, it is not statistically significant

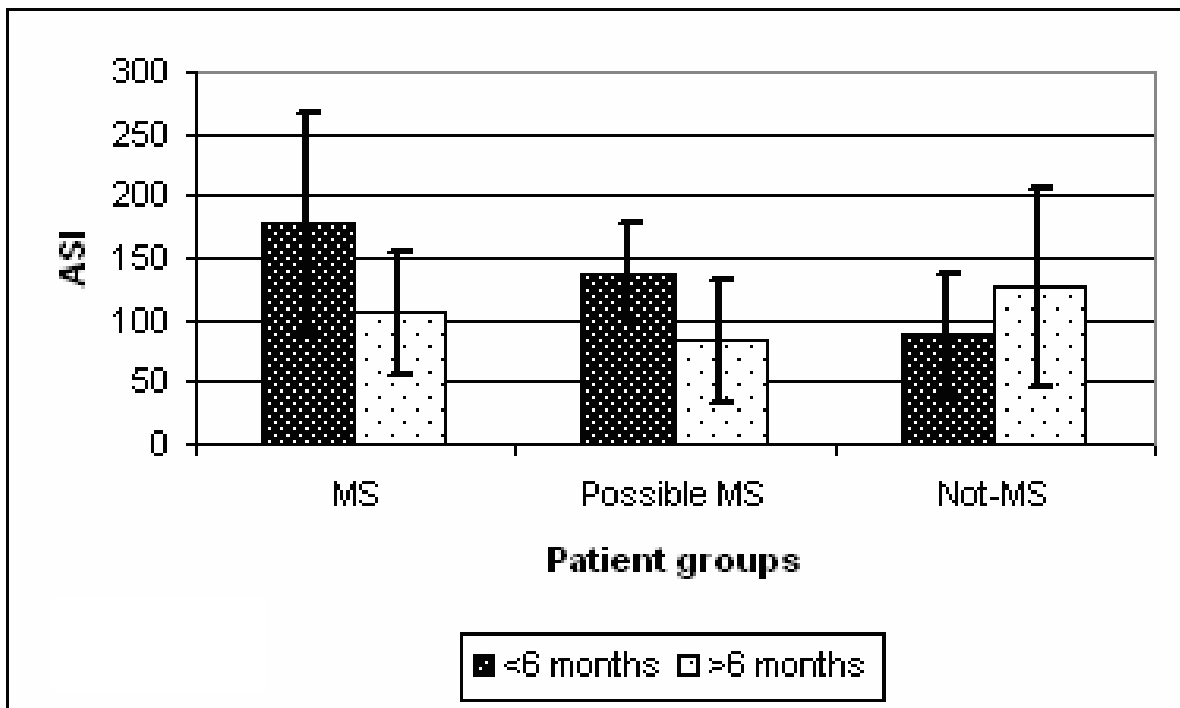


Therefore, ASI is the most sensitive indicator of amplitude abnormality for each group, with the mVEP sensitivity for amplitude loss in detecting optic neuritis of 92.6%, 92.0% and 100% in “not-MS”, “possible-MS” and “MS” groups respectively.

There was no correlation between the type of amplitude scotoma (diffuse, central, paracentral or hemifield) and the MS risk classification, ($p > 0.6$, Kruskal-Wallis ANOVA).

The initial inflammation of optic neuritis causes loss of VEP amplitudes, which have been shown to then recover after the acute episode [17]. Thus, to look at the effect of acute inflammation, the amplitude was also analysed with respect to the phase of ON. Subjects within each group were separated into 2 categories: acute phase of ON (less than or equal to 6 months) and the post-acute phase (over 6 months since diagnosis). A 6-month time frame also allows for any effect of retrograde degeneration of retinal ganglion cells to have fully developed as has been seen on pattern electroretinography (PERG) [158]. Figure 5.2 shows the change in average ASI for each of the 3 groups. Though the ASI scores were higher (indicating amplitude loss) in “MS” and “possible MS” groups acutely, the post-acute eyes in both groups showed a trend towards recovery ($p < 0.05$, Kruskal-Wallis). The ASI, and thus amplitudes, did not normalize in either group. The trend was opposite (but not significant, $p = 0.2$, Kruskal-Wallis) in the “not-MS” group with higher ASI scores in the post-acute eyes.

Figure 5.2: The average Accumap Severity Index (ASI) score for patient groups in the acute (≤ 6 months) and post-acute (> 6 month) phase of ON at the time of testing, shows a decreasing trend in those with “MS” and “possible MS” ($p < 0.05$ Kruskal-Wallis), but a trend to worsen in those classified as “not-MS” ($p = 0.2$, Kruskal-Wallis).



C Latency findings

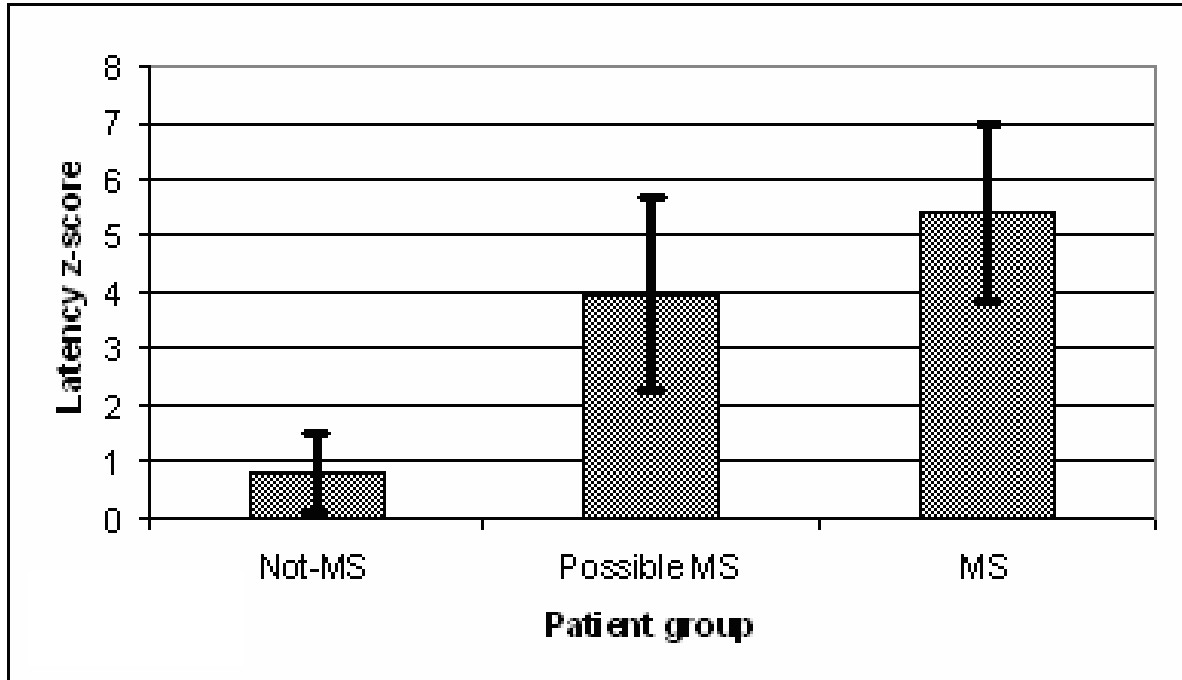
While there were only small amplitude variations between the three groups of ON patients, the latency differences were more apparent. Latency deviation abnormality was found in 100% of eyes with “MS”, 68.0% of those with “possible MS” and only in 33.3% of “not MS” eyes (Table 5.2). Within the “possible MS” group, latency asymmetry analysis increased the percentage of abnormal eyes detected. For those with “MS” there was less latency asymmetry, possibly due to either increased sub-clinical optic neuritis or MS plaques located within the optic radiations. Therefore, combination of latency deviation and latency asymmetry produced abnormality in 33.3%, 76.0% and 100% of “not MS”, “possible MS” and “MS” groups respectively.

	Not-MS	Possible MS	MS
Latency deviation	33.3%	68.0%	100%
Latency asymmetry	25.9%	76.0%	66.6%

Table 5.2: The percentage of eyes within each patient classification group with abnormalities of latency deviation and latency asymmetry.

The quantitative analysis of the latency deviation for each group, based on the sectoral mVEP waveforms, revealed a statistically significant difference in the latency z-scores between each group, ($p < 0.0001$, ANOVA) with z-scores of: 0.5 +/- 0.6 in the “not MS” group, 3.8 +/- 1.9 in the “possible MS” group and 5.8 +/- 1.0 in the “MS” group (see Figure 5.3).

Figure 5.3: The average latency z-scores for each patient classification group are significantly different ($p < 0.0001$, ANOVA).



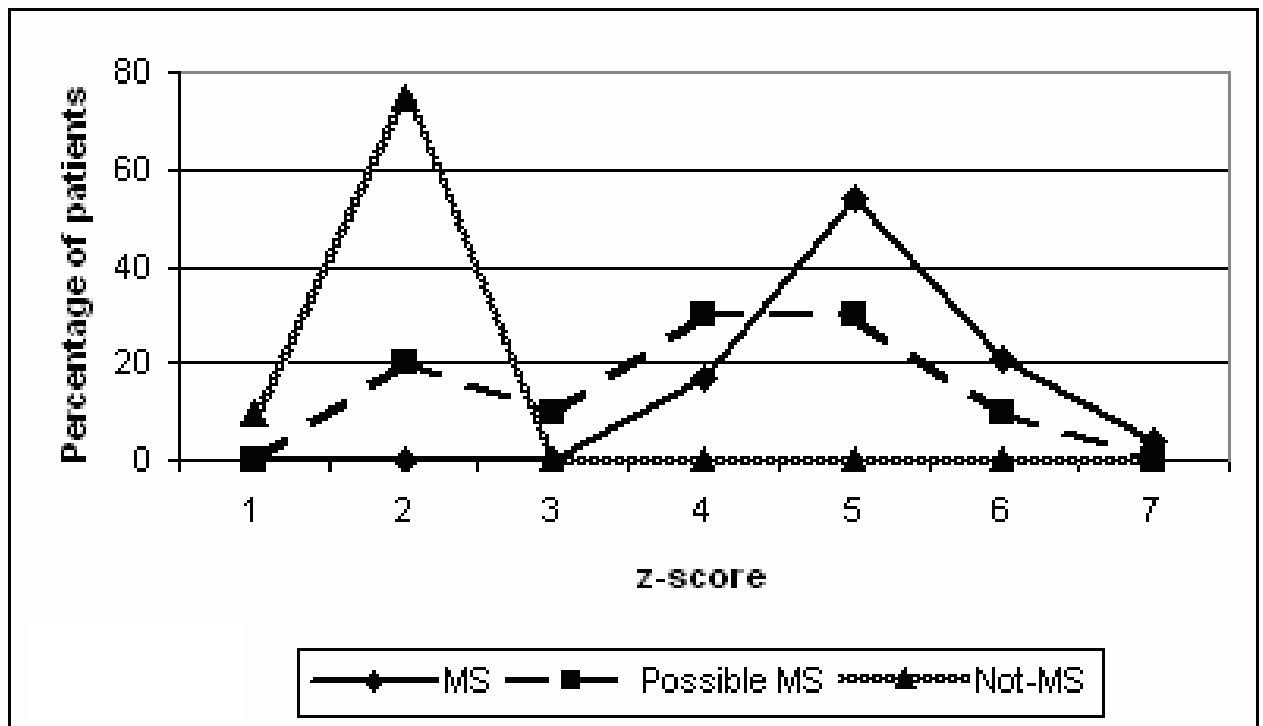
The average latency z-scores for the control group were -0.25 ± 0.76 . There was no statistically significant difference between these latency z-scores and those of the “not-MS” group ($p=0.99$, student T-test).

To evaluate the distribution of the latency z-score abnormalities within each of the patients groups, a histogram was plotted showing the percentage of patients with each z-score (Figure 5.4). There was a clear separation of the latency z-score distributions between the “not-MS” and the “MS” group. All of the “not-MS” patients had latency z-score of 2 or less (which is within 95% confidence interval for normal population).

On the other hand, within the “MS” group the majority of eyes had a z-score of 5 or greater. Therefore the sensitivity based on sectoral z-scores provided a much better separation between “not MS” and “MS” groups than detection based on the definition of a scotoma, demonstrating an abnormality in none of “not-MS” eyes but in 100% of the “MS” group.

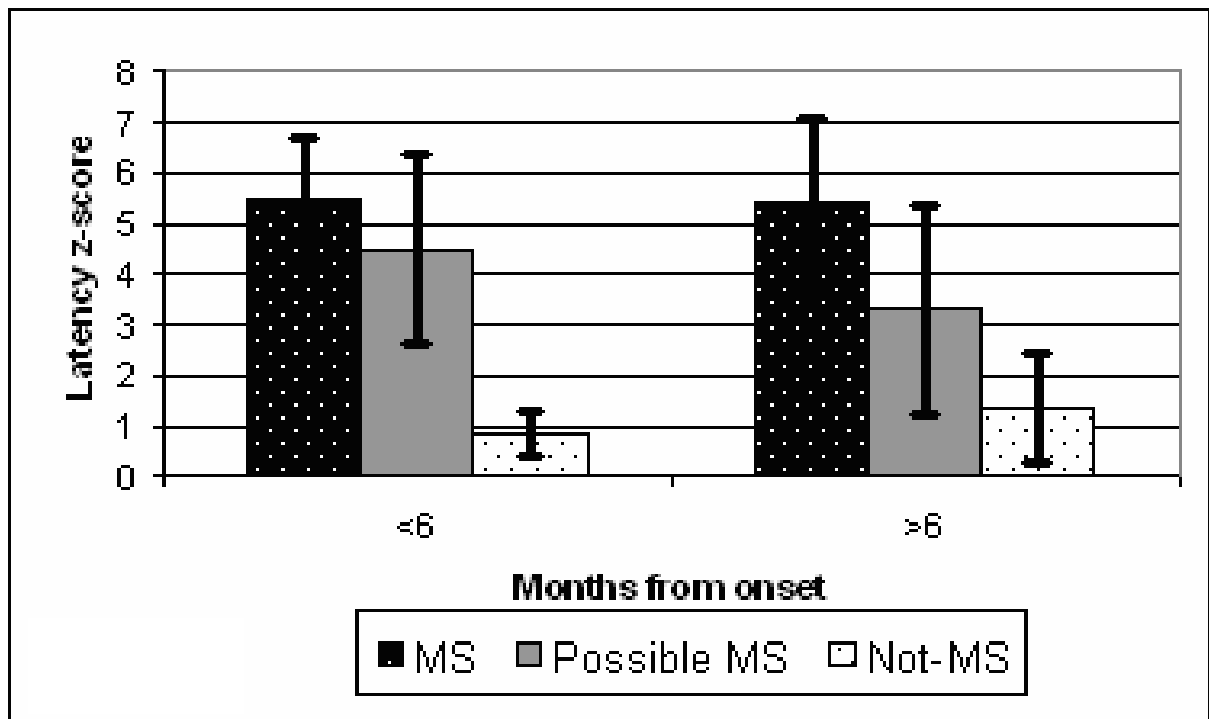
The “possible MS” group included some patients with a normal latency, while others exhibited a high z-score, indicating possible heterogeneity within this group.

Figure 5.4: The overall distribution of latency z-scores within each classification group of patients, shown as a percentage of the total number of patients within that group. The “not-MS” and “MS” groups are clearly separated. The “possible-MS” group contains some patients with normal z-scores, and others with elevated z-scores.



During the acute phase of ON the inflammatory process leads not only to significant VEP amplitude reduction, but also to moderate conduction delay [82]. Following the acute phase, inflammatory delay is resolved, thus ongoing latency delay can be attributed to demyelination [17]. Therefore, in order to separate the effect of inflammation and demyelination on the latency of mVEP, patients were again divided into acute (≤ 6 months since onset) and post-acute (> 6 months since onset) sub-groups. In acute sub-group, there was a statistically significant difference between the latency z-scores of “not-MS” and “MS” groups, ($p < 0.0001$, Kruskal-Wallis). Those groups remained separate and practically unchanged in the post-acute phase ($p < 0.0001$, Kruskal-Wallis). The latency in the “possible-MS” group, however, slightly (but not significantly, $p = 0.23$, Kruskal-Wallis) improved and while it was significantly different only from the “not-MS” group in the acute phase ($p = 0.0005$, Kruskal-Wallis), but not from “MS” group ($p = 0.46$, Kruskal-Wallis), it demonstrated significant difference from both “not-MS” ($p = 0.016$, Kruskal-Wallis) and “MS” ($p < 0.005$, Kruskal-Wallis) groups in the post-acute phase. (Figure 5.5)

Figure 5.5: The average latency score for patient groups in the acute (≤ 6 months) and post-acute (>6 month) phase at the time of testing, shows a clear separation between the “MS” and “not-MS” groups at both time points. The “possible-MS” group is closer to the “MS” group in the acute phase, though is significantly different from both “MS” and “not-MS” groups in the post-acute phase, ($p < 0.005$ and $p = 0.016$, Kruskal-Wallis).



The distribution of latency z-scores was then analysed in acute (Figure 5.6) and post-acute (Figure 5.7) sub-groups. In both sub-groups there was a very clear separation between the “not-MS” and “MS” groups as demonstrated in previous analysis. However, the pattern of z-score distribution in “possible MS” group was quite different between two sub-groups. While in acute phase all but one patient had delayed latency of varying severity, in the post-acute sub-group, the distribution was clearly bimodal. The first peak of latency z-score distributions (45% of patients) fell within the normal limit and corresponded to the “not-MS” group. The remaining 55% of patients had a latency z-score of greater than 2, with the majority forming a second peak in distribution at a latency z-score of 6, this time corresponding to the peak of the “MS” group latency z-score distribution.

Figure 5.6: The distribution of latency z-scores for patients tested during the acute phase (≤ 6 months) of ON for each classification group of patients, shown as a percentage of the total number of patients within that group. The “MS” and “not-MS” groups are clearly separated, the “possible-MS” group has patients with varying scores.

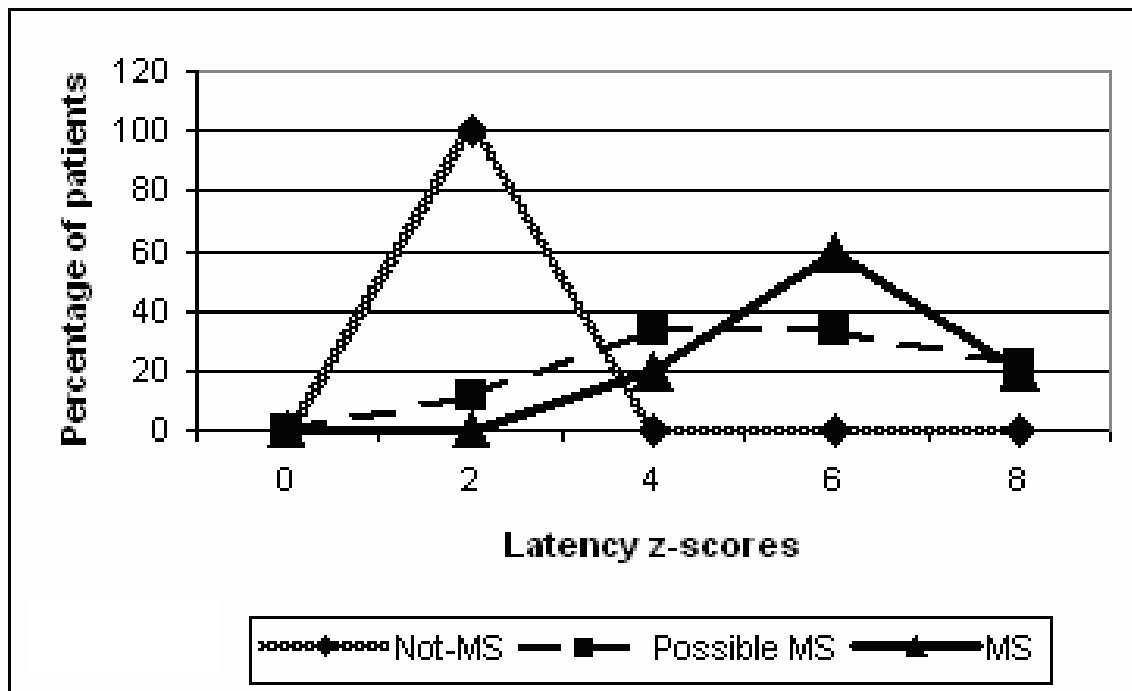
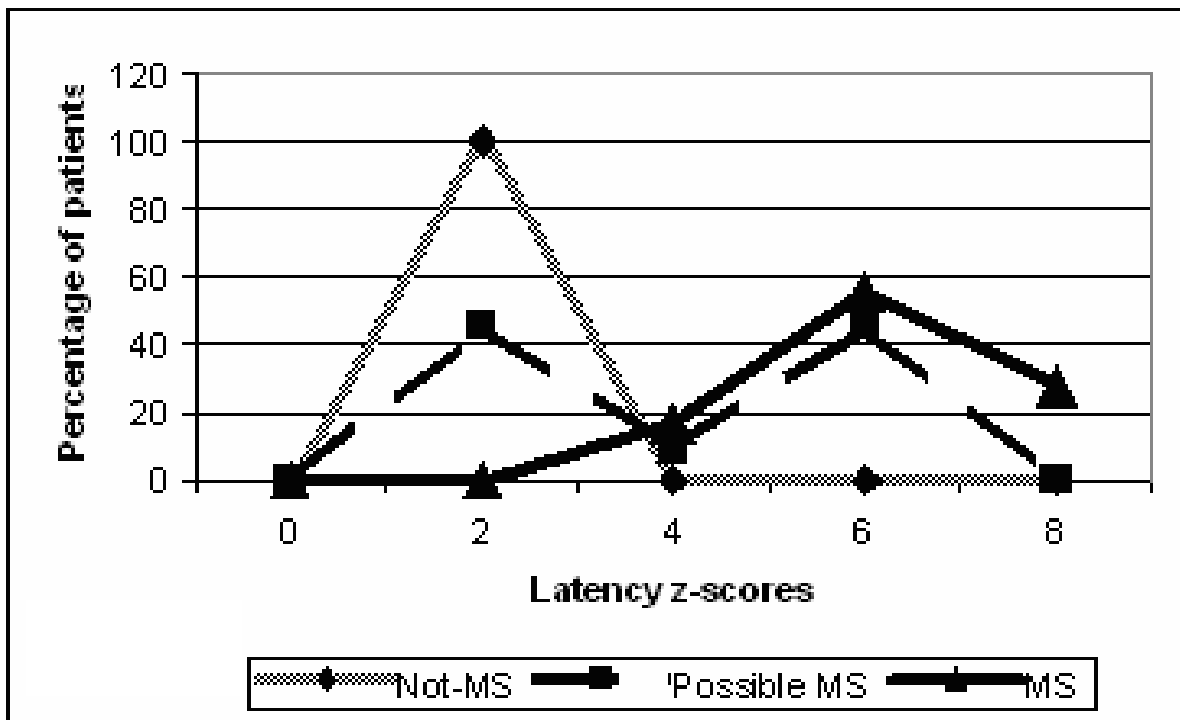


Figure 5.7: The distribution of latency z-scores for patients tested in the post-phase (>6 months) of ON for each classification group of patients, shown as a percentage of the total number of patients within that group. Again, the “MS” and “not-MS” groups are clearly separated, though the “possible-MS” distribution curve is bimodal, with peaks corresponding to those of the other two groups.



To determine if this bimodal distribution in the post-acute “possible-MS” group was due to the MRI white matter lesion load the MRI scans were re-examined, with no trend apparent between the two sub-groups of patients.

To test for the significance and interaction of other confounding factors that might influence latency differences, such as age, sex, treatment and the number of ON recurrences, a multiple linear regression (MLR) analysis was performed. An equation to describe the latency z-scores and confounding factors for each group was determined. On subsequent linear regressions, the factor with the least significance was removed from the equation and the effect of this alteration was analysed. There was no significance or interaction due to any of the confounding factors ($p < 0.01$; $F = 167$; $r^2 = 0.86$, MLR). Following the removal of confounding factors the Kruskal-Wallis test and post-hoc analyses were repeated. This demonstrated that all groups were still significantly different from each other, ($p < 0.01$ respectively).

D Discussion

This is the first study that has examined the sensitivity of the multifocal VEP technique in a large number of patients with optic neuritis classified according to the McDonald criteria. The separation of optic neuritis patients into groups with “MS”, “possible MS” and “not-MS” provided a basis from which to analyse amplitude and latency results that, to our knowledge, has not been used with full-field conventional VEP studies.

Previous VEP studies on inflammatory ON and MS have often reported conflicting results, which to some extent may be explained by lack of consistency in the classification used. Some studies have divided the patients into acute mono-symptomatic optic neuritis (AMON) or optic neuritis as part of MS [57, 70]. Other studies have chosen the Poser criteria [17, 71-74], or studied possible MS versus probable MS combined with definite MS [75-78]. However, not all patients in these MS studies had a documented episode of optic neuritis. Thus, Manguiere reported 80% abnormality in those with MS,

compared to 50% abnormality rates in those with possible or probable MS [75]. Similar findings were also documented with abnormality levels in those with MS (with or without a history of optic neuritis) between 68-100%, and those with possible or probable MS between 50-70% [75-78]. No study reported a significant difference between the groups.

Studies reporting amplitude abnormalities range from 17% to 100% in MS alone [79, 80], and 78% to 100% in retro-bulbar ON alone [57]. When comparing amplitude differences between groups, either larger amplitudes in patients with MS versus those with AMON (at 2 months post ON only) [74], or no significant difference between groups [57, 81] was found.

The literature describing latency changes in optic neuritis patients does not give a clear picture regarding latency recovery in conventional VEPs. Halliday described that latency delays may persist for many years after acute optic neuritis [55] and several other studies of both cross sectional [81, 82] and longitudinal design [83, 84] have shown long term latency reductions. Garrick et al found that patients either worsened (12%), recovered (38%) or the latency remained unchanged (50%) [85]. Brusa studied 31 patients, 22 with isolated optic neuritis and 9 with MS, no significant difference in latency analysis or visual function was found between the two groups [81]. However, 2 studies found that the mean latency was significantly shorter in patients with acute mono-symptomatic optic neuritis (AMON) when compared to those with clinically definite MS at all testing times between acute onset and one year [57, 74].

A Medline literature search did not reveal any VEP studies that have used the McDonald criteria to classify optic neuritis patients. The McDonald criteria is believed to be the most sensitive in defining the risk of MS based on clinical presentation and MRI findings [30, 159] and is recommended by the International Panel on the Diagnosis of MS [30]. It separates patients with a clinically isolated syndrome suggestive of MS, such as ON, into 3 groups: not-MS, possible MS and Multiple Sclerosis.

While the current study analysed the overall sensitivity of mVEP in the detection of ON, it also related the trends seen in the mVEP amplitude and latency findings to the patients' McDonald classification.

i. Amplitude analysis in patients grouped according to the McDonald criteria

All 3 groups had an amplitude abnormality in the majority of cases. Amplitude abnormalities were detected in 85.2% of “not-MS” eyes, 88.0% of those with “possible MS” and 100% of “MS” eyes. There was no significant relationship found between mVEP amplitudes and the group to which the patient belonged. Although a trend for worsening ASI scores from “not-MS” to “possible MS” and to “MS” did appear, larger patient numbers are required to make the statistical analysis more robust.

In both the “MS” and “possible MS” groups the mVEP amplitudes showed larger values (as indicated by a decreased ASI) in post-acute phase, which corresponds well with conventional VEP results indicating amplitude improvement after initial inflammation is resolved [17]. This trend was not demonstrated in “not MS” group, which may to some extent be explained by patient follow-up practice within the neurology clinic. In those patients classified as “not-MS” (normal MRI and no neurological symptoms), only those with ongoing visual loss, secondary to optic atrophy for example, are likely to continue with follow-up beyond a year. This could account for the relatively larger amplitude defect seen in not-MS patients in the post-acute phase of ON. A 3 year longitudinal study of not-MS patients with acute ON is in progress, to assess if these trends are applicable to individuals.

ii. Latency analysis in patients grouped according to the McDonald criteria

While there was no significant trend seen in amplitude analysis, there was a significant difference between the rates of latency abnormality (based on the definition of a scotoma) for each of the three groups: 100% for “MS”, 76.0% for “possible MS” and 33.3% for “not-MS”. The level of abnormality (based on sectoral z-scores) was also significantly

different between each of the 3 groups. No trend towards latency recovery was seen in this cross-sectional view of the MS population. Patients who were classified as “not-MS”, on the other hand, did not have latency delays at any time period post-ON. Thus even those with acute inflammation, thought to cause a conduction block did not have significantly different latency values compared to the normal population. Any effect of retrograde degeneration of retinal ganglion cells, as shown on PERG studies of ON [158, 160-164], did not affect latency results in the “not-MS” group.

In those patients with “possible MS”, latency delays in the acute group were apparent and did not differ significantly from those with MS. However, in the post-acute group a bimodal distribution of latency z-scores was discovered. No confounding factors were seen that could explain this bimodal distribution. Therefore this separation within the “possible MS” group could represent a heterogeneity based on an individual’s severity of demyelination or a failure of remyelination. It could then be hypothesized that those with longer latency delays have experienced demyelination, a hallmark for MS, whereas those with normal latency values have a different pathological cause for the ON and white matter lesions on MRI. Whether these patients continue to mirror the “not-MS” and “MS” groups in terms of clinical course and disease progression, as well as latency scores, needs to be investigated. It must be noted that even those patients with a McDonald classification of “not-MS” at the time of presentation with ON, still have a 6% chance of developing MS within one year, and a 20% chance within 5 years [157].

When test results can lead clinicians to alter patient management they are of particular value. If this latency pattern does reflect future clinical course, then the mVEP could provide a means of stratifying those with a greater risk of future MS in the early post-acute stage of ON from all of those with white matter changes on MRI. The study by Jacobs et al indicated that interferon-beta-1a when commenced after the first episode of ON in patients with 2 or more lesions on MRI (comparable with the possible MS group in this study), resulted in a 44% reduction in the conversion rate to MS at 18 months [35]. Thus there is an incentive for early treatment of ON patients with a higher risk of future MS. However many clinicians are reluctant to start all patients on treatment, with the

accompanying side-effects and complications, given that a certain percentage of them will never develop MS. If the longitudinal studies do reflect these results and confirm the hypothesis, then those patients with higher latency z-scores should be started immediately on treatment aimed to lower progression rates to MS while those with lower latency z-scores should continue to be monitored.

E One year follow-up of “possible-MS” patients

In the cross-sectional study (sections A to D) 64 subjects with optic neuritis (ON) were classified according to the McDonald criteria for Multiple Sclerosis (MS) as either “not-MS”, “possible-MS” and “MS”. Each patient then underwent multifocal visual evoked potential (mVEP) testing and latency analysis was performed. It was found that latency values are significantly different between the three groups of patients ($p < 0.001$, ANOVA). The “possible-MS” patients were shown to have a bimodal distribution of latency z-score results: some patients had no latency delay (mirroring results from the “not-MS” patients) whereas other patients had long latency delays similar to those already diagnosed as “MS”. It was hypothesized that delayed mVEP latency in a “possible-MS” patient could represent an increased probability of conversion from “possible-MS” to “MS”. Thus the purpose of this follow-up study was to determine if mVEP latency delay in those with “possible-MS” is associated with increased risk of progression to clinically definite MS.

i. Methods

68 subjects with ON were enrolled and classified according to the McDonald criteria as either “not-MS” (17 subjects), “possible-MS” (29 subjects) or “MS” (22 subjects). Each patient had a confirmed diagnosis of ON (unilateral visual loss, afferent papillary defect, pain on eye movement). Patients with any other ocular pathology were excluded. All patients had completed a 3 day course of 1g per day, intravenous methylprednisolone and

a 2 week oral taper of steroids. The mean time from onset of ON to testing was 11.9 months (range 15 days - 4 years).

29 ON patients at least 6 months from the onset of their ON were categorized as “possible-MS”. Multifocal VEP was performed using the AccumapTM (ObjectiVision, Sydney, Australia) at the start of the study. The latency z-scores were determined, and group was divided into those with delayed mVEP latency results, and those with normal results. A consultant neurologist reviewed all patients over a one year period with repeat clinical examination and MRI scans. Those patients who converted to definite MS, as per the McDonald criteria were recorded.

ii. Results

All 22 patients with definite “MS” demonstrated significant latency delay (average z-score=5.8+/-1.0, range 3.9 – 7.6). All patients classified “not MS” had normal latency (average z-scores=0.5+/-0.6, range -0.7 – 1.6). The “possible MS” patients were shown to have a bimodal distribution of latency results: some (7 patients) had no latency delay, similar to the normal controls and “not MS” patients, whereas others (22 patients) had long latency delays, similar to those with “MS”.

Over one year, 8 patients from “possible MS” group progressed to definite “MS”. All 8 patients had latency delay on initial mVEP testing. Thus 36.4% of those with mVEP latency delays progressed to MS, whereas none with normal latency from the same group progressed. This difference in conversion rates was statistically significant ($p=0.03$ Chi-squared). None of the patients in the “not-MS” group converted to MS.

Basic statistical analysis does not reveal any significant difference (treatment received, time from onset of ON) within “possible MS” group between these 8 patients and the remaining 21, however numbers are too small for robust analysis. Longer follow-up and larger patient numbers are required to determine if these results are consistent over time.

iii. Discussion

As MS is typically diagnosed in the third and fourth decades, it results in significant functional and work-related disability, in what should be the most productive years of life. The 10 year data from the Optic Neuritis Treatment Trial (ONTT) has shown that the overall risk for MS following ON is 38%, but that this increases to a 56% risk in those with one or more typical lesions on the baseline MRI (“possible-MS”) (19). However in the CHAMPS study of 383 patients with ON and 2 or more MRI white matter lesions characteristic for MS, being ovoid or periventricular, it was found that weekly intra-muscular interferon-beta-1a injections reduced the cumulative probability of developing definite MS from 50% to 35% (20). In a more recent analysis of the CHAMPS data, it was found that by using more stringent MRI criteria for eligibility as set out by Barkof (9 or more white matter lesions with at least one gadolinium enhancing lesion) the treatment with interferon conferred a 66% reduction in risk for MS over 3 years (21). Thus, in order to better target this treatment clinicians need to be able to distinguish those patients at the highest risk of future MS from all those who presented with ON and MRI changes.

Studies using the MRI evidence of dissemination of lesions in space and time, as set out in the McDonald criteria, have shown that the new McDonald criteria lead to more than double the number of patients with a diagnosis of MS at one year, compared to the use of the Poser criteria (22, 23). In addition, the American Academy of Neurology guidelines on MRI report that once alternative diagnoses are excluded at baseline, the finding of 3 or more white matter lesions of T2-weighted MRI is a sensitive predictor (>80%) of the subsequent development of MS (24). This pilot study in optic neuritis patients suggests for those patients with abnormal MRI findings (who still do not fulfill the McDonald criteria for a diagnosis of MS) that a sectoral latency delay on mVEP may correlate to an increased rate of progression to MS over the following year compared to the patients with normal mVEP latencies. However, the authors do acknowledge that even in patients diagnosed as “not-MS” with normal MRI scans, there is still a small risk of MS (25).

The potential for earlier recognition of those more likely to develop MS, from patients with an initial MRI scan suggestive but not diagnostic for MS, would allow clinicians to closely monitor this subset of patients with a view to more targeted prescription of interferon-beta-1a. This finding represents a new development in the diagnosis of MS as mVEP is non-invasive, inexpensive and can be performed as part of a routine clinic.

Chapter 6

Long term mVEP follow-up of optic neuritis patients: Acute optic neuritis to 12 months post diagnosis

A Methods

In order to determine the ability of mVEP to detect and track changes associated with ON, patients with acute ON were recruited from tertiary hospital emergency departments. Multifocal VEP assessments of amplitude and latency changes were performed. This test was performed during the acute phase of illness at 1 month from diagnosis, 3 months, 6 months and 1 year.

The full testing protocols are to be completed 2 years from the time of diagnosis. However, this thesis will only be able to report the preliminary results, up to and including the 12 month tests.

All patients underwent testing after completion of a three day course of 1g intravenous methylprednisolone.

Twelve patients have been enrolled in the study, and have completed their 12 month test. The average age is 28 +/- 5 years (range 17 – 34). At the start of testing there was one patient with MS, 7 with “possible-MS” and 6 considered as “not-MS”. Over the course of the year, 3 patients converted to a definite diagnosis of “MS”. Results were analysed based on the patient’s diagnostic category at one year.

As discussed in Chapter 5, the best indices for analysis of follow-up studies, based on low test-retest variability are overall amplitude deviation based on ASI and sectoral latency values. Thus, for this study, only these parameters were examined though sectoral

latency was analysed using latency z-scores rather than absolute values, as these vary from sector to sector.

B Results

i. Amplitude

For eight of the patients there was a steady trend towards recovery of the ASI (Figure 6.1, patient 1). In two of these patients a recurrence of the optic neuritis was detected at the one-year test, and for both cases this corresponded with a recent viral illness (Figure 6.1, Patient 3). This recovery was seen in all the MS classification groups. However in the two cases of optic atrophy (both “not-MS”) the ASI did not show a major trend towards recovery (Figure 6.1, Patient 2). The ASI has remained well over 150 in both cases, with a dense central scotoma and any minor improvement in the ASI coming from slight recovery of the peripheral visual field. Two patients had near normal ASI scores at the first test, therefore the recovery, though present, was minor (Figure 6.1, Patient 4). One patient showed a trend to worsening ASI scores, and was diagnosed as having “MS” 9 months post ON.

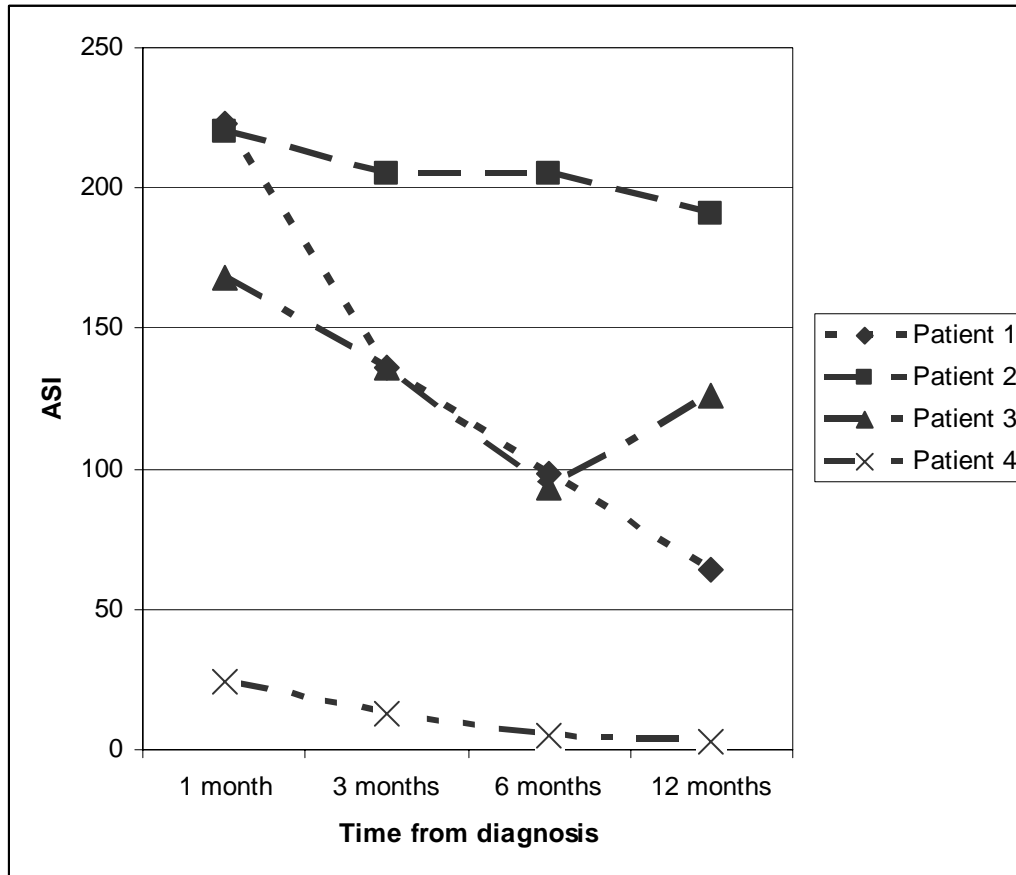


Figure 6.1: Examples of ASI results for four patients at the different test times. Patient 1 – steady recovery. Patient 2 – optic atrophy, with no recovery. Patient 3 – recurrence of optic neuritis at one year. Patient 4 – minimal abnormality at onset, full recovery.

There is a statistically significant difference in the ASI between all 4 visits when examining the results from the group as a whole ($p < 0.001$, ANOVA with Huynh-Feldt correction). However, on post-hoc comparison, there was no statistically significant difference between the ASI scores at the 3 month test and the 6 month test, $p = 0.21$. Improvement in the ASI overall from the acute one month phase to 3 months post diagnosis was 19.4%. Between the 3 month and 6 month tests there was an average improvement of ASI of 17.5%. This trend continued between the 6 month and one year tests, again with an average improvement of 19.2%. The magnitude of change in ASI score per month was -5.6 with a standard error of change of 0.98 , with 95% confidence interval between -7.6 and -3.6 . With an estimated normal fluctuation of ASI results in optic neuritis patients of 9%, this indicates clinically significant ASI improvements in

these patients. However, when looking at the ASI changes in those patients who developed optic atrophy, the fluctuations were all of <10% magnitude. Thus these changes may have just been normal testing variation.

There is no significant difference between the ASI results on each visit for the different MS classification groups, (p=0.66, ANOVA). When examining the rate of change of ASI between visits, there was no significant interaction due to the MS classification group. In other words, the difference seen in ASI between visits is not dependant or different between the MS classification groups, (p=0.27, ANOVA). See Table 6.1.

	1 month	3 months	6 months	12 months
MS	189 (+/- 62)	142 (+/-36)	120 (+/- 77)	83 (+/- 79)
Possible-MS	131 (+/- 72)	101 (+/- 50)	72 (+/- 36)	61 (+/- 51)
Not-MS	121 (+/- 97)	110 (+/- 92)	100 (+/- 92)	90 (+/- 88)

Table 6.1: Mean Accumap Severity Index for each patient group at each visit, with standard deviation.

Interestingly both “MS” patients showed a large improvement of ASI from >100 to <50 in each case, on commencement of interferon.

ii. Sectoral latency

The changes to sectoral latency z-scores did not follow an apparent trend over time. In 4 of the patients classified as “not-MS” the latency z-scores were less than 2 and remained within the normal range over the year (Figure 6.2, Patient 5). In one “not-MS” patient the amplitudes in the affected eye were too small to make an accurate latency assessment. In three of the “possible-MS” patients latency z-scores indicated latency delay at the one month and 3 month tests, however 2 had returned to within the normal range at the 6 month test (Figure 6.2, Patient 6). In four of the patients, latency z-scores were abnormal on the first test, and have become steadily higher over the course of the year, indicating an increasing latency delay. Two of these patients were diagnosed with progression to

MS during this one year period, based on a second clinical neurological episode (Figure 6.2, Patient 7). At the time of the final test, neither patient had been commenced on disease modifying drugs. Of the two other patients, both showed latency delay at the first test, which was increased at the three month test. Both patients were diagnosed with MS in this time, commenced on a form of interferon treatment, and latency z-scores have returned to normal in one case, and 2.5 in the other (normal <2). This may indicate that the treatment is allowing remyelination to take place within the MS lesions (Figure 6.2, Patient 8).

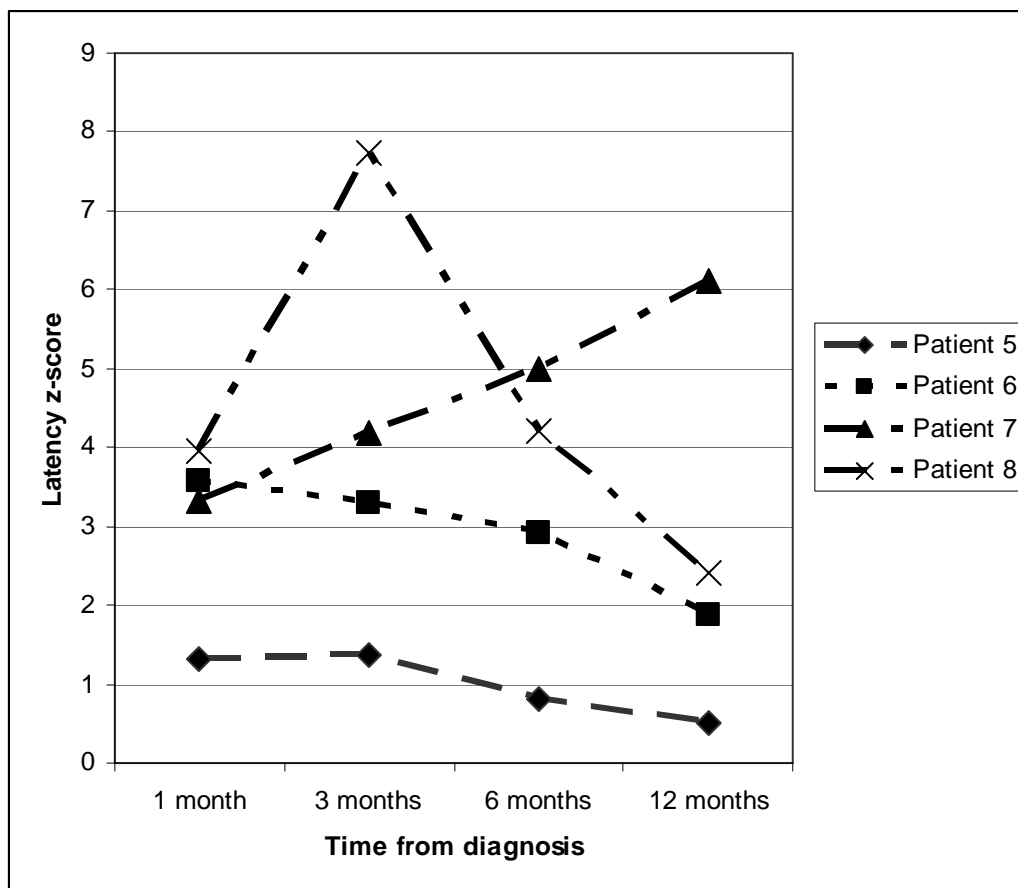


Figure 6.2: Latency z-score changes over time in example patients. Patient 5 – normal latency throughout. Patient 6 – latency recovers to normal. Patient 7 – steady rise in latency prior to diagnosis of MS. Patient 8 – rise of latency, followed by latency recovery after commencing MS treatment.

On statistical analysis there was no overall statistically significant difference in the latency z-scores between the four test times, ($p=0.29$, ANOVA, Huynh-Feldt correction), see Table 6.2. However there is a significant difference between the latency z-scores at each test time, between the patients based on MS classification, ($p=0.001$, ANOVA). Based on post-hoc comparisons, there was a statistically significant difference between the z-scores of patients classified as “not-MS” and the “possible-MS” and “MS” groups ($p=0.003$ and $p=0.001$ respectively). The magnitude of change of the latency z-score per month was calculated at -0.06 with a standard error of 0.04 , and was not statistically significant ($p=0.1$, ANOVA). A change in latency values between tests was not dependant on the MS classification groups, ($p=0.5$, ANOVA).

	1 month	3 months	6 months	12 months
MS	3.55 (+/- 0.37)	5.11 (+/-1.9)	4.27 (+/- 0.5)	3.47 (+/- 1.9)
Possible-MS	4.19 (+/- 1.2)	3.24 (+/- 1.22)	3.9 (+/- 2.3)	3.18 (+/- 1.9)
Not-MS	1.31 (+/- 0.8)	1.43 (+/- 0.8)	1.27 (+/- 0.9)	0.68 (+/- 0.7)

Table 6.2: Average latency z-scores with standard deviations, for each patient classification group, at each test time.

C Discussion

These results show that it is possible to use the mVEP to detect and track the changes of optic neuritis over time. Secondly, that the changes seen as part of the normal disease process are outside the limits of normal test-retest variability. This is consistent with the conclusion drawn by Hood et al. based on 3 ON patients, that mVEP can be used to track disease recovery [110].

The results from this ongoing study were consistent with the results from our own cross-sectional series of ON patients as there remained a statistically significant difference between the latency z-scores of patients based on MS risk classification. Those patients classified as “not-MS” had normal latency z-scores, those with “MS” showed latency

delay. Those patients classified as “possible-MS” showed a combination of normal and delayed latencies, and that these latencies can and do normalise by the 6 month post diagnosis test. This further confirms the bimodal distribution of latency z-scores seen in the cross-sectional study. Furthermore, of the patients who converted to MS during the course of the year, all had delayed latencies. Interestingly, one patient who converted to MS was initially classified “not-MS” by the McDonald criteria, having normal brain and spinal MRI scans, and no evidence of another neurological episode other than ON. However, throughout testing she had high latency z-scores indicating latency delay, and the delay worsened rather than improved over the tests. This did not fit with our original assertions in Chapter 6. Nine months post ON, she developed parasthesia in her left hand, and MRI now reveals demyelinating lesions in the spinal cord, that were not initially present. According to the McDonald criteria, she now has “MS”. This further strengthens our case for delayed mVEP latencies at the 6 month test indicating an increased risk of progression to MS.

The most common findings on conventional VEP studies are of a markedly reduced and delayed VEP during the acute phase, with increasing amplitudes during the recovery phase, but that latency values do not normalise [55-57] and often remain constant for up to 5 years [165]. Results from this study were consistent with these findings with respect to amplitude recovery.

In a two year study looking at retest scores on cVEP in patients with ON, it was noted that in post-hoc t-tests there was a significant decrease in mean VEP latency between 3 months and 6 months, but at all other time points, the changes were not significant [81]. Our study revealed that there was little overall change in the mean latency values. It would be possible that in a study with mainly “possible-MS” patients, and it is not clear in the study quoted which MS classification was used, that the recovery of latency between 3 and 6 months that we have described, could become statistically significant.

Of more interest is the response of the latency z-scores in the patients commenced on interferon therapy. Both patients showed a marked recovery of latency delay following

initiation of treatment. This effect needs to be examined further, and could potentially be used to track the functional success of new MS treatment options. However, there is increasing evidence from pathological studies that following an episode of demyelination in MS, that new myelin is formed [166]. There are a number of other reports indicating that cVEP latency delays have been shown to return towards normal [70, 167]. Brusa et al, described the mean P100 latency becoming significantly shorter at 3 years compared to 6 months. However they found that there was no difference in amplitude between 6 months and 3 years [70]. It was hypothesised that remyelination caused the latency recovery, whereas axonal loss complete at the 6 month test was permanent, thus no change was seen in amplitude results. Our results do not entirely support this, with ongoing amplitude recovery seen between the 6 month and one year mark.

The recovery of latency seen in those with “possible-MS” in our study could reflect an improvement in the conduction block caused by inflammation, which in turn causes latency delay, or it could represent the return of a functioning myelin sheath. Given that remyelination has been shown to occur in MS plaques [168], the worsening latency values seen in the patients who progressed to MS, could be due to subclinical development of lesions within the optic radiations. Longer follow-up of our patients is required before full comparison with the study by Brusa et al can be made.

In conclusion this study reveals that amplitude recovery continues for at least up to one year in all MS classification groups. Latency values on average do not change over time, but are significantly different between patient groups. Latency values may increase, as was seen in some patients who developed MS, or improve as seen in some “possible-MS” patients and those “MS” patients started on treatment, or remain the same as seen in the “not-MS” group. This study will be continued for up to 2 years, to track these changes further.

Chapter 7

Improving Multiple Sclerosis detection: Neural network analysis of mVEP waveforms to improve trace quality

A Introduction

As discussed in Chapter 5, in order to make the mVEP more useful in a clinical setting, the test-retest variability should be low enough to allow for analysis of disease progression. Latency variability is affected by the amplitude of the mVEP waveform, which is in turn affected by the signal to noise ratio. Thus one of the aims of this study was to increase the signal to noise ratio allowing for more accurate diagnoses and patient follow-up.

Despite the best efforts of the technician there may be trace contamination with ECG noise, alpha rhythm or muscular activity resulting in a low signal to noise ratio. The noisy traces may be used by the software in the final diagnosis producing false negative or false positive results. The current mVEP device (Accumap V2.1) tested in this study uses a combination of filters to separate a true VEP trace from noise, and algorithms to improve the signal to noise ratio (SNR). These filters are often not adequate to recognise and remove a large noise spike within the first 250 ms of a trace from a real VEP trace. Thus a value for amplitude and latency may be assigned to what is essentially noise, and this may lead to an inaccurate diagnosis. This has implications for future progression analysis in following cases of optic neuritis and the misdiagnosis of latency changes is of particular importance in multiple sclerosis.

Artificial neural networks are computational programs based on the neuro-biological architecture of a feed-forward system of artificial “neurons” that learn from example data. [169] Each artificial neuron receives a number of inputs signals, either the original data or outputs from other neurons. Each input is associated with a connection weight. The neuron processes the weighted sum of these inputs and subtracts this from the sum of

the neuron threshold. This is then passed through a non-linear transfer or activation function to produce the output for that neuron. Input data passes sequentially into hidden layers of neurons, progressively activating certain neurons. Layers of these neurons are interconnected to produce pre-determined outputs allowing for a powerful non-linear modelling and advanced pattern recognition. [170]

Artificial neural networks require sample data to make the internal connection between the inputs and output relationship and thus learn a structure in the data set. During this training period, the specific network algorithm works to adjust the neuronal activations, to produce the desired output for each set of inputs. Connection weights are sequentially modified to reduce output errors. If training has been successful the network should then be able to process unseen input data to produce an output, often a classification of the input data.

A recent review article makes the case for the importance of neural networks in electroencephalogram (EEG) processing, with many positive results indicating a place for these systems in the future of medicine [171]. Artificial neural networks have been used to detect specific graphic elements of an EEG with correct pattern recognition results varying from 69-89% [172, 173]. Neural networks have been used to detect disease states in the EEG such as schizophrenia[174], Huntington disease, Parkinson's disease[175] and Alzheimer's disease[176] from normals. [177]

In the field of ophthalmic diagnostics neural networks have been used to classify and diagnose with varying degrees of success. By presenting a system with retinal images one group achieved 79% recognition accuracy of retinal haemorrhage over 160 images.[178] A back propagation network with three hidden layers has been used to identify glaucomatous visual field progression from sequential AGIS scores. The network predicted probability of progression with 86% sensitivity and 88% specificity. [179]

A group in Italy has combined a series of networks for the classification and analysis of adult EEGs. Firstly, a network classifies 2-second sections of EEG traces based on morphology. A second system then analyses the temporal sequence of the morphologies to provide a diagnosis based on the whole trace.[180] It thus seems feasible to create a series of networks that can classify mVEP waveforms from noise, and use a second network in the diagnosis of disease states. This experiment represents preliminary results of the first stage of this process – classification of mVEP waveforms.

B Methods

i. Data

Accumap™ traces can be described by a sequence of 500 data points with an interval between points of 2.2msec. The 500-point trace array data from all segments of the mVEP recording in 28 eyes (14 subjects, 1624 traces) was exported to an Excel spreadsheet. A variety of recordings were chosen to represent good traces and noisy traces from various contaminants (alpha rhythm, ECG, muscular activity).

Graphic representations of each trace (0-1100ms) were then examined by 2 experts for classification of the trace as either a good / true representation of a VEP waveform (which was normally within a range of 30 to 100 points) versus a noisy trace where the VEP signal was impossible to distinguish from the noise. These results were combined to produce a consensus agreement for the classification. This resulted in a data set with 1076 true traces, and 548 noisy traces. It was decided to train the network with a majority of true traces due to the varying morphologies of true traces. Fig 7.1-7.3. The network had to be able to recognize both positive and negative deflection waves as true.

Figure 7.1: Normal mVEP waveform.

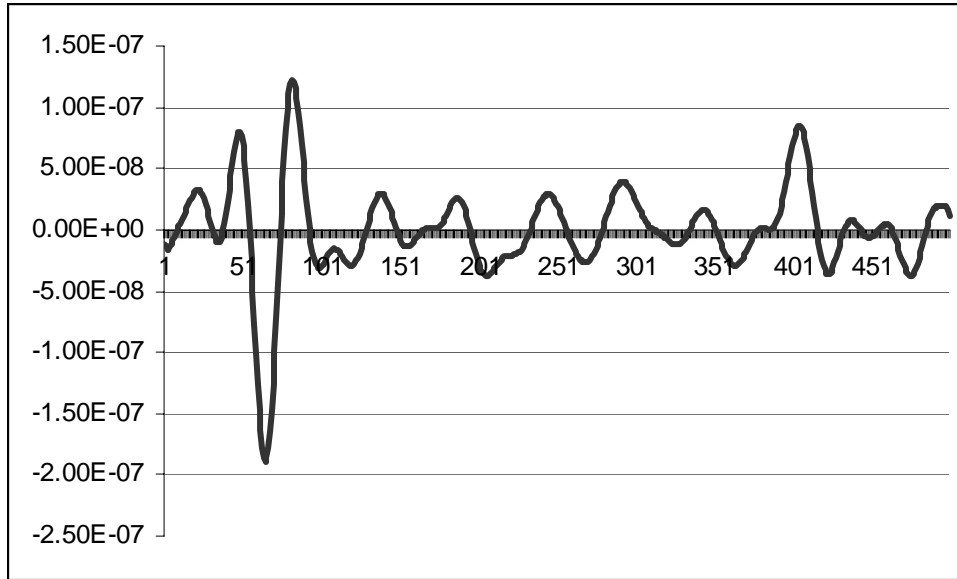


Figure 7.2: Small mVEP waveform with noisy trace background.

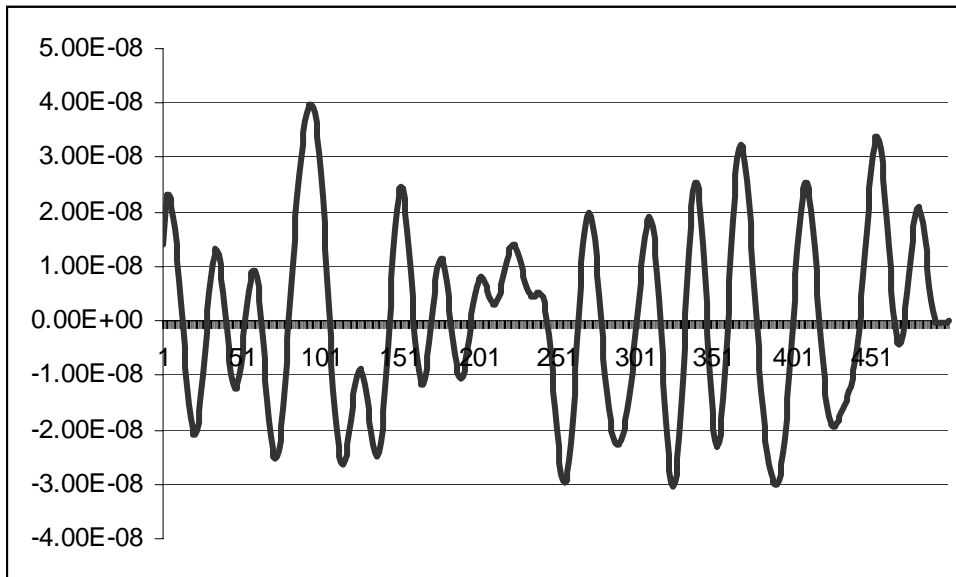
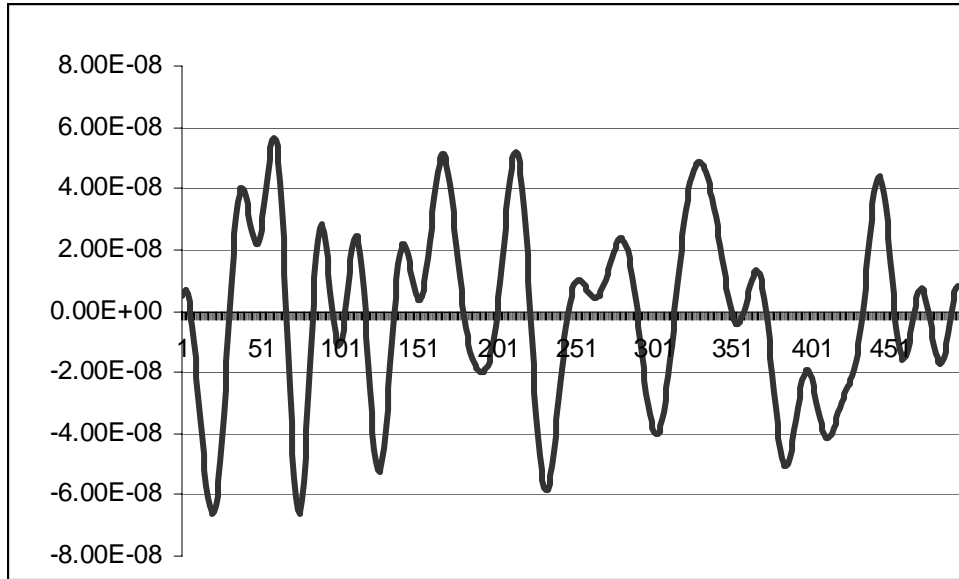


Figure 7.3: Noise signal.



Statistica Neural Network software (version 7, StatSoft, Inc., OK, USA, with recent updates from their homepage) was used to produce the neural networks.

The input data was presented to the software in a variety of formats. Firstly, the raw data points for each case were used, as a 100, 250 and 500-point trace. Fig 7.4, 7.5, 7.1.

Secondly, a root-mean-squared (RMS) and averaging function was applied to make the waveforms all of positive inflection. Fig 7.6. The averaging function, took an individual point and averaged it with the surrounding 4 points, to create a new value for that point.

This smoothed out the effects of the RMS. Our aim was to reduce the variety of input patterns considered as a “true” trace. Finally we entered the normalized amplitude of the raw data to eliminate incorrect classification of smaller amplitude waveforms.

Figure 7.4: First 100 data points displayed of figure 7.1

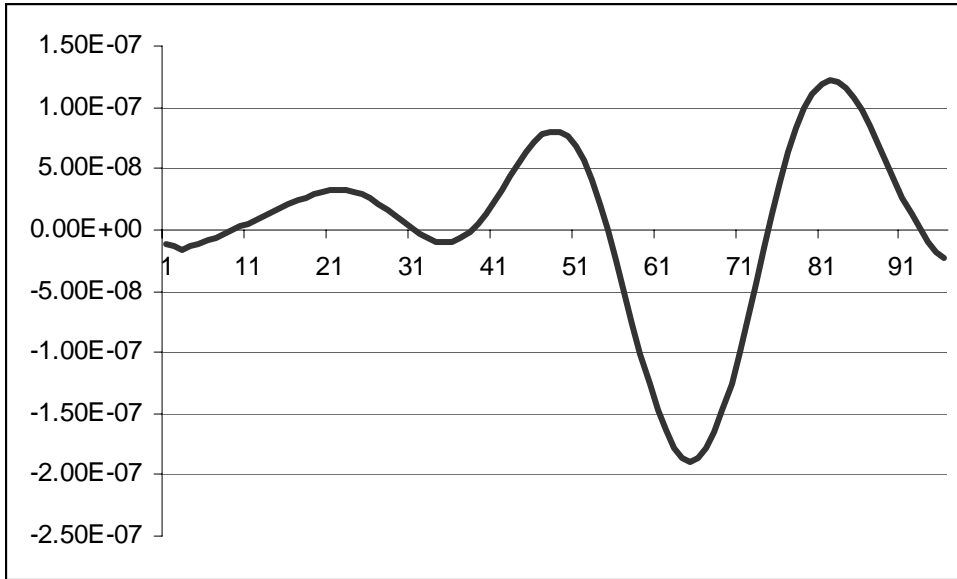


Figure 7.5: First 250 data points displayed of figure 7.1

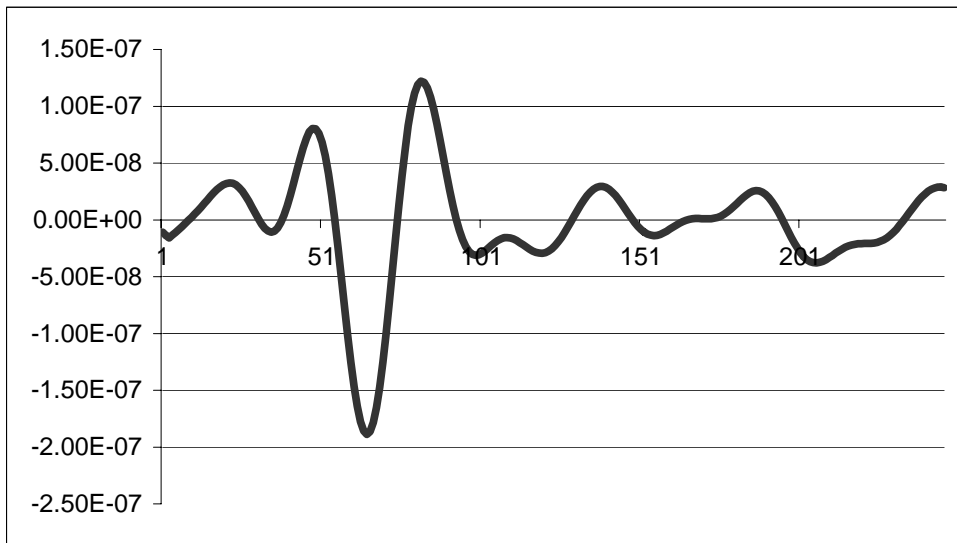
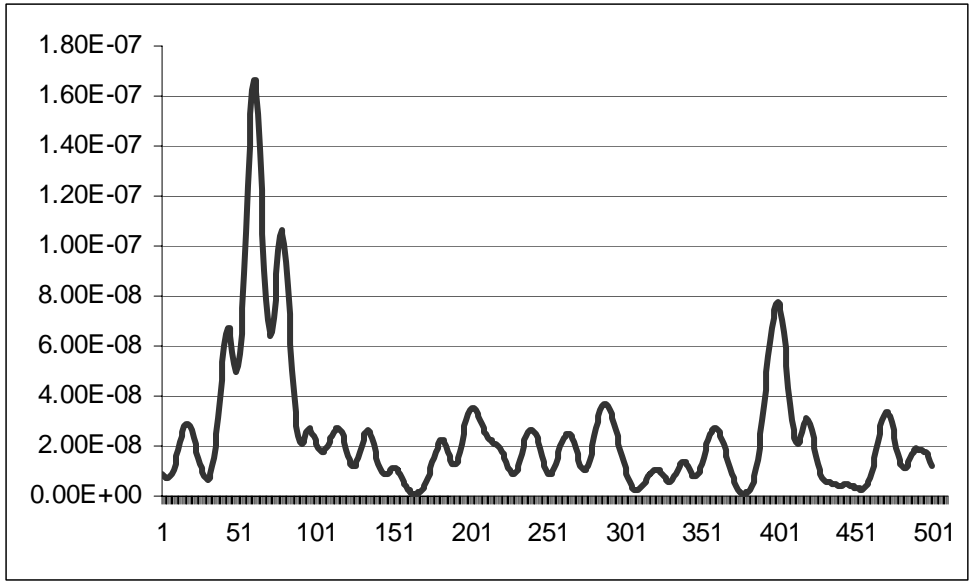


Figure 7.6: Root mean squared and averaging function applied to figure 7.1.



A total of 1624 traces were classified and presented in the above-mentioned forms to a variety of network types for training. Half of the traces were randomly selected as the training data for each run. Another 25% of the cases were used for verification, to keep an independent check on the progress of training and where further refinement of the system is performed. Finally the last 25% (406 traces) were used only to test the system accuracy at the end of a training sequence.

ii. Networks

The Statistica neural network software allows alterations of different network types for refinement and classification improvement. Multilayered Perceptron network and Radial Basis Function network were used in this study.

Multilayered Perceptron

The most popular network type is a multilayered perceptron (MLP) that trains with a back-propagation algorithm and a second-order algorithm such as the conjugate gradient descent. The system progresses through a series of epochs, where each training case is submitted to the network. Desired and actual outputs are compared, the error and the error surface gradient are used to adjust the connection weights, and the next epoch begins. The network is initially random, and training stops either when the error is acceptable, the error stops improving or a given number of epochs have passed.

This network can have many layers, with the input and output layer sandwiching any number of hidden layers. The number of neurons within these hidden layers can also be varied by the network architect. We started with a 3 layer network, with the number of hidden neurons equal to half the sum of the number of input and output units. Both back-propagation and conjugate-gradient-descent training algorithms were tried. The StatsSoft program chose automatic learning rates, momentum and stopping conditions.

Iterative experiments were conducted with each type of learning algorithm and each configuration of the network. The best network was retained (in terms of selection error).

On each experiment where under-learning, or inadequate results occurred, more neurons were added to the hidden layer, and when this did not improve results significantly, extra hidden layers were added. When over-learning occurred, i.e. the selection error started to rise, hidden neurons and hidden layers were removed.

After many iterative experiments, the best network configuration for each type of data entry was kept. We then re-sampled and generated new networks with the same configuration to see if better results could be obtained. As the main purpose of this experiment was to improve trace recognition, only the results for the best networks are described. The exact architecture of each network is not described.

Radial Basis Function

These networks are also based on a 3-layered system – one input layer, one hidden layer with radial units and a linear output layer. Whereas MLPs divide the pattern space up using lines, radial basis function networks (RBF) use spheres. The MLP neuron uses a linear vector based on the input and connection weight. The RBF neuron responds to the distance of an input value from the “centre” represented by the radial unit, before passing this through the activation function. The response of each unit is a Gaussian (bell-shaped) function. Since these functions are non-linear it is possible to model any function shape with only one hidden layer. This makes for ease of network design, as the number of hidden units is the only variable. These networks also train faster due to the limited complexity. However, this radial approach is very localized, thus these networks often perform poorly when extrapolating from data that deviates drastically from the training data.

Many RBF networks were tested each with different numbers of radial units, centres and deviations. Centres were assigned using sub-sampling (random), K-means algorithm, isotropic (same for all units) and K-Nearest Neighbour algorithms. The K-Nearest Neighbour algorithm sets each unit’s deviation individually to be equal to the mean distance to its nearest neighbours. Thus deviations are smaller in tightly packed areas of

space, preserving detail, and higher in sparse areas of space. This makes sense for our data, as more points are required to represent the true trace at the start of the data, and fewer points are needed to describe the subsequent noise.

The software automatically trains or optimizes the output layer using a standard linear optimization. Again, the exact configurations of networks are not described, as a different network was designed for each type of input data.

C Results

Based on network architecture described in the methods, the more input points, the more case examples are required to adequately train a system. Thus in an attempt to make the database size more manageable and speed up the training and running times of the networks, smaller subsets of the collected data were tested first. When the networks were presented with only the first 100 or 250 data points (Table 7.1, first and second data columns), the waveform made up about 50% and 20% of the total trace respectively, thus not giving the network enough information to distinguish noisy traces. Systems trained with 250 points had a high sensitivity but a very low specificity. After trying inputs based on the first 100 points, 250 points and the full 500 points we found that specificity results were best with the full 500-point traces (Table 7.1, third data column).

Table 7.1: Comparisons of the sensitivities and specificities of Radial Basis Function networks (RBF) and Multi-Layered Perceptron networks (MLP) for different input value types. (100= first 100 data points only used. 250= first 250 data points only used. 500= all data points used. RMS ave = root mean squared function applied to all points, with consecutive groups of 10 points averaged, to give 50 data points. Normalised = 500 data points normalised to between -1 and +1)

		100	250	500	RMS ave	normalised
RBF	Sens	81	85	87	83	90
	Spec	69	65	88	84	89
MLP	Sens	54	55	52	54	61
	Spec	43	44	49	53	54

Initial poor results were thought to be caused by the fact that true waves can have different morphology depending on the point in the visual field and the channel from which it came. Thus we performed a root mean squared (RMS) function in a way similar to analysis used previously [121] and averaged the data over the surrounding 4 points to smooth out the trace. The traces were then all of a positive inflection only, therefore reducing waveform variability. This did not however significantly improve results (Table 7.1, forth data column).

The main improvement of classification rates came from normalizing the waveform amplitudes. Often small but true waveforms of <100nV, as seen in disease states like glaucoma and optic neuritis were misclassified by the networks as noise, resulting in low specificities. Detection of those waveforms can be enhanced by normalization. We divided each data point by the difference between the maximum and minimum value for data points of each case. Thus the waveforms were normalized between +1 and -1. Once the networks were presented with normalized data, specificities soon approached the sensitivity of the network (Table 7.1, fifth data column).

Having started with both MLP and RBF network architectures, we found early success with the RBF using normalised full 500-point data sets, as seen above. Thus we focused on the RBF method, though it is likely that similar results could have been found with the MLP following rigorous architectural modification. The initial RBF network was taken and additional training and adjustment was performed to improve it further. All further results are based on this network.

To improve an RBF network the StatsSoft online textbook (www.stastsoft.com) recommends to pruning the input data based on low sensitivities, or a large radial spread for an individual input point, a function performed automatically by Statistica if desired. We found that if real wave was present (which is normally within an interval of 30-100ms) it could either be a large positive or large negative inflection (i.e. large radial spread), which was then pruned by the system. Thus the network was pruning out the biggest key to detecting a true waveform – the networks became very specific but lacked sensitivity. When the network was forced to retain and calculate each of the 500 input points, sensitivity improved. There are other ways to prune network data, which may have worked better in this case, however all initial attempts at data pruning only served to reduce the sensitivity of the network.

We designed a radial basis function network that retained all 500 input points, and used 100 hidden units to trigger a classification output for each trace. The receiver-operated curve value is 0.977.

Each network can be re-trained using the data set to improve performance. The risk is that the network will become over-trained and too “fitted” to the training data. When this occurs, the training error improves, but the test error worsens. There is no evidence of over fitting of the data as the percentage of correct classification is not markedly different between the training data and the test data. (Table 7.2)

Table 7.2: Training, verification and test results for the final version RBF.

	Training set		Verification set		Test set	
Classification	True mVEP	Noise	True mVEP	Noise	True mVEP	Noise
Total	551	261	256	150	269	137
% Correct	94.2	92.7	92.2	94.0	92.9	93.4
% Wrong	5.8	7.3	7.8	6.0	7.1	6.6

One of the most important functions of this network in clinical application will be to detect noisy signals and thus remove them from the final report and diagnostic functions. We simulated a completely noisy trace by testing a subject without cross-correlation between the visual stimulus and EEG sampling. While there was no real signal present, the Accumap system filters allowed 40/116 traces in each eye to be used and reported an amplitude and latency in those segments. The remaining 76 the system classified as “no trace”. This is a diagnostic accuracy of 65%. The same traces were then run through the neural network. Of the total 116 traces, the network classified 113 as noise. Therefore, the use of the network increased diagnostic accuracy up to 97.4% in this case. (Table 7.3)

Table 7.3: Current mVEP filters vs. neural network at correctly identifying all 116 traces as noise.

	Current filters	Neural network
Total	116	116
Correct	76	113
Wrong	40	3
Diagnostic accuracy	65%	97.4%

D Discussion

There are two major misclassification problems that can arise from a noisy mVEP trace. Firstly, the system can miss a true scotoma (false negative case). In a case of true pathology the amplitude of the mVEP signal can be greatly reduced or even completely absent. However, a noise spike if it is situated within the window for analysis (70-220msec) may mistakenly be assigned amplitude and latency values. In this situation pathology can be underestimated or missed entirely. Normally the averaging that occurs over the course of the test will eliminate much of a noise, but this may not always happen particularly in patients with high alpha rhythm for example. This type of misclassification has implications for detection of small visual field defects and for monitoring of the progression of diseases such as glaucoma and optic pathway tumours, where it is vital to recognize low amplitude signals that may represent an increase in the size of the scotoma.

Secondly, there may be a real mVEP signal that is missed by the system, and thus a scotoma may be incorrectly diagnosed (false positive case). For example, slight reduction in amplitude caused by early pathological changes (particularly in an upper field where signal is known to be smaller) may lead to a situation when signal is lost in the background noise. True signals of normal amplitude can also be missed if there is a prolonged latency as in the case of multiple sclerosis.

By normalizing the waveform amplitudes we were able to train a neural network to accurately classify a real mVEP waveform and therefore separate them from noise with a sensitivity of 92.9% and a specificity of 93.4%. This neural network system of classification will be of great benefit in trace recordings that have a known small signal to noise ratio. Often in the elderly patients or those who are anxious the mVEP signals are noisy due to an excess of muscular electrical activity in the neck and shoulders. Young male subjects often have very high levels of alpha rhythm from the start of the recording that cannot be overcome by any amount of concentration or prompting from the technician. Patients with a slim body habitus may have high levels of ECG contamination of the mVEP traces.

Neural networks can be used to identify true mVEP traces from noisy traces, following adequate data pre-processing. By integrating the neural network into the mVEP software system to check all the final trace recordings before amplitude and latency calculations are performed the incorrect diagnosis of scotomas may be prevented and accuracy of progression analysis may be improved. There are plans for future versions of the software to carry such trace recognition software.

Chapter 8

Conclusion

Visual field assessment and visual evoked potentials have long been used in the field of neuro-ophthalmology. Thus it seems important to begin to expand the role of multifocal VEP, which provides both perimetric and electrophysiological assessment of the visual system, into the field of neuro-ophthalmology. The results from this study have taken the concept of multifocal visual evoked potentials from an objective perimeter used in glaucoma detection, through to further its potential as a diagnostic machine in optic neuritis and Multiple Sclerosis.

In the discussion of results in Chapter 3, the sensitivity and specificity of the mVEP in detecting the presence of optic neuritis was compared to the conventional VEP. Though direct comparisons with the conventional VEP results were not done in the patients tested in this study, it was possible to show that the mVEP results were at least comparable to published journal reports on conventional VEP devices. By comparing the mVEP results of optic neuritis patients, from a broad cross-section of times from diagnosis, with normal patients and patients with other causes for visual disturbance, it was possible to show that this technique could detect optic neuritis, and differentiate it from other conditions.

Comparison of supplemental visual tests such as visual acuity and colour vision, with previous optic neuritis studies, such as the Optic Neuritis Treatment Trial, showed that our cohort of patients was representative of the typical optic neuritis patient.

Furthermore, this study showed that the mVEP amplitudes were correlated with visual acuity, brightness perception, relative afferent papillary defect and abnormalities in the Humphrey Visual Field perimeter. This indicated that the mVEP could provide reliable information on the functional state of the visual pathway.

Having shown that the mVEP can detect the presence of ON, and relate to the severity of functional changes in typical optic neuritis patients, it was necessary to show that the

mVEP could be useful in a clinical setting with repeat tests. Studies in press have shown that the mVEP amplitude results have a variability of 30-40%, which can be reduced to 20% with further data analysis [149]. Therefore the study described in Chapter 4 extended this analysis to examine latency variability in normal patients. It was shown that based on the 58 test points used in the mVEP, that there was a large variability of latency values on a point by point basis. This variability increased with eccentricity from the fixation point, and with signals of smaller amplitude. However, sectoral latency analysis proved to have a test-retest variability within a range that was practical within a clinical setting. Though the sectoral averaging causes some loss of information, this is sacrificed in return for reproducibility. Thus only sectoral latency values were assessed in further studies in this thesis.

Given the reported variability of perimetric results in optic neuritis patients [153] and central fixation shifts can cause poor mVEP reproducibility (a possible problem in optic neuritis patients with a central scotoma) [151], the mVEP was tested for reproducibility in optic neuritis patients. It was shown that amplitude as measured by ASI was reproducible within 9%, though only in patients with severe defects. Amplitude deviation scotoma shape was also reproducible, however the severity of that defect was not. Sectoral latency values were as reproducible as normal patients. Thus it was reasonable to continue testing the clinical use of the mVEP in optic neuritis patients.

The significance of optic neuritis lies in its association with Multiple Sclerosis. Thus for any new optic neuritis test to be of clinical use, it would be desirable if it could give some reflection of the pathology underlying the optic neuritis. Only MRI scans have been shown to be able to predict progression to MS, following an isolated attack of optic neuritis. In Chapter 5 the role of the mVEP in classification of optic neuritis patients, according to their MS classification was examined. The McDonald criteria was chosen for the classification of the optic neuritis patients, as it incorporates the latest MRI data in the diagnostic criteria, and does not use conventional VEP results (except in the diagnosis of primary progressive MS). It was shown that the amplitude results varied little between ON patients classified according to the McDonald criteria. However there was a clear

and statistically significant difference between the sectoral latency z-score results. Those with a diagnosis of MS all showed significant latency delay. Those with a classification of “not-MS” all had normal latency values. This in itself is an interesting finding, as all conventional VEP studies report some latency delay in patients with idiopathic ON. However, once the latency values were averaged over the whole visual field, this delay was no longer apparent, even in the acute phase of ON. Of even more interest was that patients with the intermediate classification of “possible-MS” showed a bimodal distribution of latency delays 6 months after diagnosis of ON. Of these patients 55% had some form of latency delay (like those with MS), and 45% had normal latency values (like those “not-MS” patients). It is known that 56% of patients with one or more lesions on MRI at the time of ON (our “possible-MS” patients), will develop MS within the next 10 years. We hypothesised that those patients with latency delay were at higher risk of progression to MS.

In order to follow-up this hypothesis 29 “possible-MS” patients were examined using the mVEP. Clinical follow-up with neurological consultation and MRI was performed over one year on these patients. Of those patients with latency delay, 32% progressed to MS, whereas none of those with normal latency delays progressed. This difference in ratios between the two groups was statistically significant ($p < 0.01$, Chi-squared). These results lend further weight to our hypothesis.

In Chapter 6 the results of the prospective study from repeated mVEP testing to optic neuritis patients from acute stages of the disease for one year are discussed. As shown in the results of Chapter 4, it was known which indices were reproducible and worth examining over a series of repeated tests. The results from Chapter 5, lead us to closely examine the relationship of the results to the McDonald MS classification. It was shown that there was a steady improvement in amplitude measurements over one year, but that no difference existed between MS classification groups. These results were consistent with some published findings on conventional VEP follow-up of optic neuritis patients, as well as our own results from earlier chapters. Though there was no overall change in sectoral latency z-scores for the year, there was a statistically significant difference

between the three groups. Confirmation of the bimodal distribution of “possible-MS” patient’s sectoral latency z-scores was made. Conversion to a full diagnosis of “MS” over the year was only seen in patients with latency delay on their 6 month test, again reinforcing our earlier hypothesis.

Finally, given that the better the mVEP reproducibility, the more use it will have in tracking changes associated with optic neuritis and MS, a system was devised to improve signal recognition. As was shown in Chapter 4, latency reproducibility increases with signal amplitude, or signal to noise ratio. Often noise within the mVEP trace can cause the inbuilt software to produce erroneous results, which will decrease reproducibility and increase diagnostic uncertainty. Thus an artificial neural network was designed that increased diagnostic accuracy of the mVEP device in detecting real mVEP signals. Such networks have been successfully used before in EEG trace recognition, and could be installed into the software of the mVEP device.

Future work following on from this thesis will involve the longer term follow-up of acute optic neuritis patients. Patients have been enrolled and consent for a 2 year study. There are also many more patients who have yet to even reach their 12 month test. This will be combined with ongoing follow-up of these patients to watch for conversion to Multiple Sclerosis. More robust statistical data needs to be collected before it is possible to say if the latency delay seen at the 6 month test does truly represent an increased risk of progression to MS. Further work will also centre on developing this technique as a means for assessing visual pathway function and recovery in response to current and new therapies aimed at modifying the course of MS, encouraging remyelination or even curing the disease.

In conclusion, by using both the amplitude and latency data provided by the mVEP, we have expanded its role into one of more than just an objective perimeter, and expanded the diagnostic field of optic neuritis and Multiple Sclerosis. The mVEP has been shown to be of use in the diagnosis and tracking of optic neuritis. There may also be a role for the mVEP in assessing the future risk of Multiple Sclerosis in patients presenting with

optic neuritis. As treatment becomes available in Australia (as it is already in the US and Europe) that can reduce the risk of Multiple Sclerosis after optic neuritis, it will be increasingly important for clinicians to be able to diagnose and monitor these two conditions.

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