TAU PROTEIN AND BRAIN DERIVED NEUROTROPIC FACTOR PROFILE IN PATIENT UNDERGOING SEVOFLURANE ANAESTHESIA

By

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ABBREVIATIONS

ASA	American Society of Anaesthesiologist
Tau	Tau Protein
BDNF	Brain Derived Neurotropic Factor
ETT	Endotracheal tube
MAC	Minimun Alveolar Concentration
ELISA	Enzyme-linked Immunosorbent Assay
HR	Heart rate
EDTA	Ethylenediaminetetraacetic acid
MAP	Mean arterial pressure
RR	Respiratory rate
POCD	Post Operative Cognitive Dysfunction
BIS	Bispectral index
MME	Mini Mental State Examination
ETCO2	End Tidal Carbon Dioxide
OR	Operating theatre
PCA	Patient Control Analgesia
GA	General Anaesthesia
SD	Standard deviation
CI	Confidence interval
IHD	Ischaemic heart disease

ABSTRAK

KESAN SEVOFLURANE KE ATAS PARAS TAU PROTEIN DAN BRAIN DERIVED NEUROTROPIC FACTOR SELEPAS PEMBEDAHAN

Objektif: Sevoflurane ialah gas yang seringkali digunakan dalam pembiusan penuh untuk memastikan pesakit berada dalam keadaan tidak sedar dan pembedahan dapat dijalankan dengan lancar. Secara amnya gas sevoflurane ini selamat digunakan untuk pembiusan pesakit namun terdapat kesan jangka panjang terhadap fungsi kognitif pesakit tersebut. Kesan penurunan fungsi kognitif ini telah dikaitkan apabila paras Tau Protein dan Brain Derived Neurotropic Factor didalam badan pesakit juga berkurangan selepas pembiusan menggunakan gas sevoflurane. Kajian ini adalah bertujuan untuk mengetahui kesan gas sevoflurance ke atas paras Tau Protein dan Brain Derived Neurotropic Factor di dalam badan pesakit selepas menjalani pembedahan yang menggunakan gas sevoflurane untuk pembiusan penuh.

Methodologi: Kajian ini berbentuk prospektif keratan rentas ‘ cross sectional study ‘ yang telah dijalankan di Dewan Bedah Hospital Universiti Sains Malaysia daripada bulan Jun 2013 sehingga Oktober 2014. Sebanyak 39 orang pesakit orthopaedic yang menjalani pembedahan menggunakan gas sevoflurane untuk pembiusan penuh telah terlibat didalam kajian ini. Sebelum pembiusan dijalankan, darah telah diambil untuk analisa penanda Tau Protein dan Brain Derived Neurotropic Factor. Ketika pembiusan dijalankan suhu badan pesakit dipastikan 36 – 37.5 °C untuk memastikan paras tau protein dan BDNF tidak terjejas disebabkan suhu yang sejuk. Sepanjang pembiusan am dijalankan menggunakan gas sevoflurane BIS digunakan untuk memastikan tahap MAC

(konsentrasi alveolus minimum) gas Sevoflurane adalah 1.5 – 2.0 dengan campuran oksigen : udara, 70 : 30. Jangka masa pembedahan adalah diantara 60 – 180 minit. Selepas pembedahan dijalankan pesakit akan di ektubasi dan di letakkan di bilik pemerhatian. Pesakit akan dilihat oleh doctor bius dan memenuhi kriteria ALDRETE dan PADS skor sebelum di hantar ke wad. 24 - 48 jam seterusnya selepas pembedahan, satu lagi sampel darah telah diambil. Asai ELISA untuk plasma Protien Tau dan BDNF telah dilakukan untuk mendapatkan paras darah untuk penanda bio asas sebelum pembedahan dan selepas pembedahan yang menggunakan gas sevoflurane untuk pembiusan penuh.

Keputusan: Purata paras Tau protein sebelum menggunakan gas sevoflurane adalah 18.63 ± SD 18.84 (purata ± SD) dan purata paras Tau Protein selepas menggunakan gas sevoflurane adalah 10.52 ± SD 18.52. Purata paras BDNF sebelum menggunakan gas sevoflurane adalah 1.63 ± SD 1.71 dan purata paras BDNF selepas menggunakan gas sevoflune adalah 1.40 ± SD 2.06. Tiada sebarang perubahan ketara dapat dilihat dari segi paras Tau Protein atau Brain Derived Neurotropic Factor sebelum dan selepas pembedahan menggunakan gas sevoflurane. Ini menunjukkan bahawa tiada kesan keatas paras Tau Protein dan Brain Derived Neurotropic Factor pesakit selepas hanya sekali menjalani pembiusan menggunakan gas sevoflurane.

Kesimpulan: Kedua-dua paras Tau Protein dan Brain Derived Neurotropic Factor tidak berubah selepas hanya sekali menjalani pembiusan menggunakan gas sevoflurane.

ABSTRACT

TAU PROTEIN AND BRAIN DERIVED NEUROTROPIC FACTOR PROFILE IN PATIENT UNDERGOING SEVOFLURANCE ANAESTHESIA

Objective: Sevoflurane is commonly and widely used inhalational agent in general anaesthesia. The sevoflurane will be used in general anesthesia to ensure patients are not aware during the operation. Generally, anaesthesia using sevoflurance is safe but some study showed that the usage of sevoflurane can cause cognitive impairment post operatively in susceptible patient. The use of sevoflurane may cause deterioration in neurocognitive function resulting from tau hyperphosphorylation and perhaps reduced level of brain derived neurotropic factor. The purpose of this study is to investigate the effects of sevoflurane anaesthesia on the level of tau protein and brain derived neurotropic factor.

Methodology: This was a cross-sectional observational study from June 2013 until October 2014 done in operation theatre Hospital Universiti Sains Malaysia (USM). 39 patients scheduled to undergo elective surgery in orthopaedic cases requiring general anaesthesia were included. Blood was obtained for baseline tau protein and BDNF before induction of anaesthesia using sevoflurane. Maintenance of anaesthesia using sevoflurane with the target minimum alveolar concentration (MAC) 1.5 -2.0 in oxygen: air, 70:30 mixture and to achieved BIS reading of 40 – 60. Throughout the surgery body temperature are maintained within normal range (T° 36- 37.5). Duration of anaesthesia was planned for at least 60 and up to 180 min. Once operation is finished, patient will then be extubated and observed in recovery room & will be discharged to the respective

wards once they met the ALDRETE & PADSS score & reviewed by anaesthetist. Blood investigation for Tau Protein and BDNF will be repeated 24-48 hours postoperatively. ELISA assay for the plasma Tau protein and BDNF were performed to obtain the blood level for these biomarkers.

Results: The mean level of pre Tau Protein was 18.63 ± SD 18.84 and the mean level post Tau Protein was 10.52 ± SD 18.52. The mean level of pre BDNF was 1.63 ± SD 1.71 and the mean level of post BDNF was 1.40 ± SD 2.06. There were no significant changes in the level of Tau Protein or Brain Derived Neurotropic Factor before or after undergoing anaesthesia using sevoflurane. Postoperatively, there were no significant differences in the level of Tau Protein or Brain Derived Neurotropic Factor after first time exposure to sevoflurane anaesthesia.

Conclusions: There were no significant differences on the level of both Tau Protein and Brain Derived Neurotropic Factor after first time exposure to sevoflurane anaesthesia.

CHAPTER 1: INTRODUCTION

For more than a half-century, anesthetics have been used in the clinical setting whilst their mechanisms of action on the brain remained unknown. The lipid theories of anesthesia (Meyer Overton rule) claimed that anesthetics produced loss of consciousness via direct interactions with the lipid bilayers of the neuronal membranes. It is well accepted now that anesthetics produce their effects on the CNS by modulating the activity of ligand- and/or gated ionic channels located on the neuronal membranes in the CNS. However, the mechanisms of anesthetic actions are complex, and considering only one ionic channel as the unique relevant target of anesthetics cannot account for the reported effects of these agents in the brain (Grasshoff C *et al*., 2005)

Anesthetics given during surgery produce changes in the patient's behavioral state by modifying brain activity via at least two mechanisms: dose-dependent global, and regionally specific, suppression of neuronal activity and disruption of functional interactivity within distributed neural networks (Heinke, W. and Koelsch, S. 2005). Neuronal nicotinic acetylcholine receptors (nAChRs) consist of different subunits, α and β, with different subtype arrangements corresponding to distinct pharmacological and functional properties. It has been demonstrated that nAChRs are involved in cognitive processes such as learning and memory and control of movement in healthy subjects. Recent data from knockout animals have extended the understanding of nAChR function. Dysfunction of nAChR has been linked to a number of human disorders such as schizophrenia, Alzheimer's and Parkinson's diseases (Hogg, R.C *et al.*, 2003)

Interestingly, there is accumulating experimental evidence that anesthetics also affect brain functions on the long term, both in a desirable (preconditioning, neuroprotection) and maybe undesirable (neurotoxicity, postoperative cognitive dysfunction, sleep-wakefulness disorders) with particular sensitivity of the elder brain. These long term actions of anesthetics are likely to be mediated by interference with cellular signaling going from DNA transcription into RNA to the post-transcriptional regulation of protein activity by phosphorylation (J. Mantz and S. Dahmani 2008)

Despite technological advances in surgery and anesthesia during the last few decades, the incidence of postoperative cognitive dysfunction remains a relatively common complication in surgical patients. After surgery, elderly patients in particular often exhibit a transient reversible state of cerebral cognitive alterations. Anesthetics administered as part of a surgical procedure may alter the patient's behavioural state by influencing brain activity. This concise report will address the scientific evidence on the relationship between postoperative cognitive dysfunctions and the most common inhalational agents currently used in anesthesia (P.K Mondal *et al*., 2009).

Postoperative cognitive dysfunction (POCD) is a decline in a variety of neuropsychological domains, especially in memory and executive function, but also is characterized by a slowing of brain processing speed. Postoperative cognitive dysfunction is a distinct entity from postoperative delirium. The hallmarks of delirium are an acute state of confusion with alterations in attention and consciousness. The literature is sometimes confusing in that the two terms are sometimes used interchangeably leading to imprecision in the discussion of etiologies and potential prevention or amelioration of POCD. Though POCD is a common finding after surgery

and anesthesia there is currently no ICD-9 code for this disorder or syndrome. Postoperative cognitive dysfunction affects both young and old who present for surgery. However, the elderly have an increased incidence of this disorder due to less plasticity in the aged brain. As the world population ages, the burden imposed by POCD will become increasingly more apparent (Szokol JW *et al*., 2010)

In 2008 a meeting of leading physicians and scientists met in Philadelphia, Pennsylvania with the purpose to study the potential relationship between anesthesia and the onset and progression of neurodegenerative disorders such as Alzheimer's disease. The consensus statement concluded that there exists sufficient evidence at multiple levels to warrant further investigations of neurodegeneration after anesthesia and surgery.

Postoperative cognitive deficits are common in adult patients of all ages at hospital discharge but only the elderly are at significant risk for long-term cognitive problems. A report by the International Study of Postoperative Cognitive Dysfunction (ISPOCD) show an international trial of elderly patients (mean age 68 years, range: 60 – 81 years) who underwent noncardiac surgery demonstrated a 26% incidence of POCD 1 week after surgery, with 10% having persistent POCD 3 months later. Meanwhile younger patients (mean age 51 years, range: 40-60 years) showed a lesser incidence at 1 week postoperatively (19%) which decreased to 6% after 3 months (P.K Mondal *et al.*, 2009). Given that the incidence of POCD is common, it is therefore important to diagnose and manage POCD by implementation of validated screening tools and pharmacological treatment.

CHAPTER 2: LITERATURE REVIEW

2.1 SEVOFLURANE

Research to develop a safe, non-inflammable inhaled anaesthetic agent began in the 1930s when chemists discovered that the substitution of fluorine for other halogens "lowers the boiling point, increases stability, and generally decreases toxicity" (I. Smith *et al.*, 1996). Sevoflurane (fig. 1) was first synthesized in 1968 by Regan at Travenol Laboratories, Illinois, while he was investigating a series of halomethyl poly fluoroisopropyl ethers. The compound was initially reported by his co-workers in 1971 (Wallin RF, Napoli MD., 1971). Development was later to be impeded by apparent toxic effects, eventually shown to be a consequence of flawed experimental design. Baxter Travenol sold the rights to sevoflurane to Anaquest (Ohmeda/BOC), who in turn sold these to Maruishi Company. Maruishi continued research and development, eventually releasing sevoflurane for clinical use in Japan in May 1990. By the end of 1993, an estimated 1 million patients had received sevoflurane (Eger EI *et al.*, 1994).

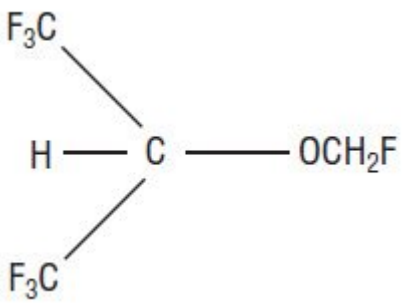


Figure 2.1: Chemical Structure Of Sevoflurane (Source : Drugs.Com)

Sevoflurane, volatile liquid for inhalation, a nonflammable and nonexplosive liquid administered by vaporization, is a halogenated general inhalation anesthetic drug. Sevoflurane, is fluoromethyl 2,2,2,-trifluoro-1-(trifluoromethyl) ethyl ether (FDA, drugs.com). Sevoflurane is nonpungent, has minimal odor, produce bronchodilation similar in degree to isoflurane, and causes the least degree of airway irritation among the currently available volatile anaesthesia. For these reason, sevoflurane is acceptable for inhalational induction of anaesthesia (Robert K. Stoelting, 2006)

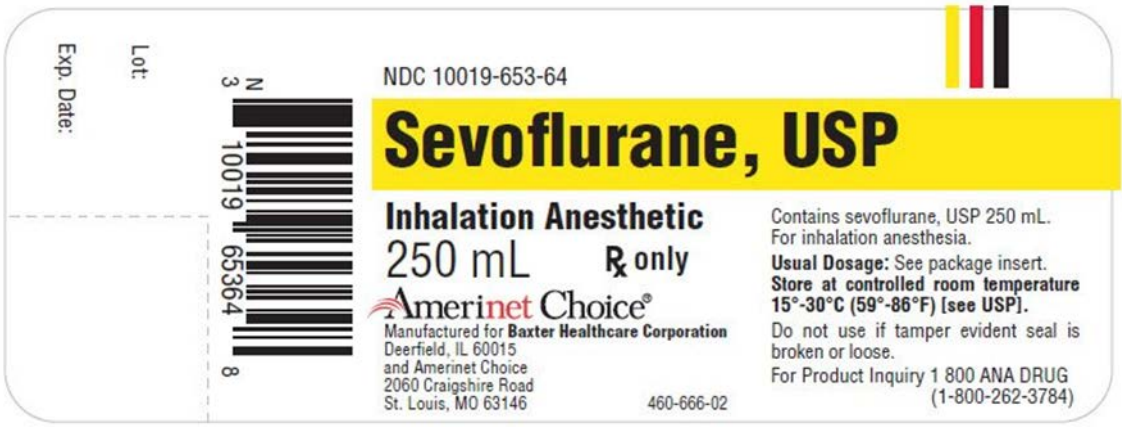


Figure 2.2: Sevoflurane Container Label (FDA Drugs.com)

Sevoflurane has little effect on normal myocardial blood flow, is a less potent coronary arteriolar dilator than isoflurane, and does not appear to cause “coronary steal” (Kersten JR *et al*., 1994). In contrast with other halogenated ethers, sevoflurane appears to be associated with a lower heart rate which helps to reduce myocardial oxygen consumption and assists myocardial perfusion (Frink EJ jr *et al*., 1992). Sevoflurane has CNS effects similar to those of isoflurane and desflurane. Intracranial pressure increases at high inspired concentrations of sevoflurane however, this effect is minimal over the 0.5–1-MAC range (Scheller MS *et al.*, 1998).

Sevoflurane is currently considered the inhalational agent of choice for adult and paediatric anaesthesia. Sevoflurane is an ether inhalation general anaesthetic agent with lower solubility in blood. The low solubility and the absence of pungency facilitate rapid mask induction; the low blood solubility also expedites "wash-out' and therefore recovery from anaesthesia (Patel, S.S., Goa, K.L., 1996). The pharmacodynamic effects of sevoflurane on the various organ systems appear to be similar to those of other commonly used halogenated ethers. Sevoflurane produces dose-dependent ventilatory depression and also reduces respiratory drive in response to hypoxia and increases in carbon dioxide partial pressure, comparable with levels achieved with other ether anaesthetics (Doi M., *et al.,* 1994)

Boiling point	58.6 °C	(at 101.325 kPa)
Density	1.517–1.522 g/cm³	(at 20 °C)
MAC	2.1 vol %	
Molecular weight	200 u	
Vapor pressure	157 mmHg (22.9 kPa) 197 mmHg (26.3 kPa) 317 mmHg (42.3 kPa)	(at 20 °C) (at 25 °C) (at 36 °C)
Blood:Gas partition coefficient	0.68	
Oil:Gas partition coefficient	47	

Table 2.1: Sevoflurane Physical Properties

The low blood:gas solubility of sevoflurane should permit rapid elimination from the CNS. When sevoflurane and isoflurane were compared as anaesthetic maintenance agents after induction of anaesthesia with midazolam and thiopentone in healthy patients undergoing elective operations, the average times from end of anaesthesia to eye opening on command were 18.6-2.0 min for isoflurane and 7.5-0.5 min for sevoflurane (Frink EJ jr *et al*., 1992). When propofol was used as the induction agent before operations lasting approximately 2.5 h, emergence from sevoflurane–nitrous oxide anaesthesia (4.1-2.2 min) was still significantly more rapid compared with isoflurane–nitrous oxide (6.7 2.2 min) (Smith I, *et al.,* 1992).

Sevoflurane was associated with faster emergence compared with isoflurane using a similar technique for gynaecological operations of shorter duration, suggesting that the use of intraoperative opioid analgesics may have masked differences between the two groups in the former study, which also involved very small numbers of patients (Quinn AC, *et al.,* 1994). Rapidly eliminated anaesthetic agents are used most commonly for day-case anaesthesia, where rapid, clear-headed recovery may allow for earlier discharge of patients.

Rapid recovery compared with halothane, and smooth and calm emergenced are also advantageous in children. For adults, the relative insolubility of sevoflurane should facilitate control of anaesthetic depth during the maintenance period, even at low gas flows. Rapid emergence from anaesthesia with sevoflurane facilitates more efficient patient turnover and may confer additional benefits after more prolonged anaesthesia (I. Smith *et al.,* 1996).

Figure 2.3: Example Bottle Of Sevoflurane

Sevoflurane is a comparatively unstable molecule. It undergoes a moderate degree of metabolism (approximately 5 %) and also breaks down in the presence of soda lime and Baralyme at elevated temperatures (Strum DP *et al.*, 1987). Both processes result in potentially toxic products. However, unlike other anaesthetic ethers, sevoflurane does not possess a CF2H group, and so (similar to halothane) does not result in the production of carbon monoxide in association with excessively dry carbon dioxide absorbants (Fang ZX *et al.*, 1995).

Sevoflurane is absorbed and degraded by both soda lime and Baralyme. When sevoflurane is mixed with soda lime, sealed in a flask and heated, a total of five breakdown products are produced. These have been designated as compounds A, B,C, D and E (Brown BR *et al.*, 1992). Concern has been raised because these products are toxin in rats. The evidence suggests that the concentration of compound A achieved in clinical practice is well below the concentration which is toxic to animals.

The effects of this drug in relation to the incidence of POCD have been investigated, but the results are not so clear. In their study on patients undergoing coronary bypass graft surgery, (Kadoi *et al*., 2007) found no relationship between POCD and the use of this anaesthetic agent. In contrast some comparative studies of sevoflurane and other volatile anesthetics, such as desflurane and isoflurane, indicate that the former seems to be associated with the worst cognitive outcomes (Kanbak *et al.,* 2005). For instance, it is clear that sevoflurane increases tau and P- tau level in the cerebrospinal fluid of surgical patients for at least 48 hours. Tau hyperphosphorylation might be a mechanistic bridge linking between anesthesia and the risk of cognitive impairment.

The effects of sevoflurane anesthesia on POCD have also been evaluated in comparison with intravenous anesthesia with propofol, highlighting how the incidence of POCD levels does not depend on the anesthetic agent used (Rohan, D *et al.,* 2005). Moreover, total intravenous anesthesia with propofol/remifentanil- shows no patient benefit over sevoflurane/fentanyl-based anesthesia in terms of recovery and cognitive functions (Magni, G *et al.,* 2005).

Although halogenated agents are thought to have limited effects on POCD, sevoflurane anesthesia has been shown to alter exploratory and anxiety-like behavior in animals with a genetically modified cholinergic system (Wiklund A *et al.,* 2008). Data showed sevoflurane promote tau phosphorylation even in normothermic conditions. Dysregulation of this phosphorylation/dephosphorylation balance involved in these key physiologic neuronal pathways may thus be impaired in sevoflurance condition (Helene L.F *et al.,* 2012).

2.2. TAU PROTEIN

2.2.1 Tau Overview

Neurons are cells with a very complex morphology that develop two types of cytoplasmic extensions, axons and dendrites. Neural transmission occurs through these processes, and therefore, any changes in neuronal morphology may affect their behavior and even produce pathological events. Indeed, it should be born in mind that the morphological differentiation of a neuron involves the extensive rearrangement of the cytoskeleton, which is responsible for maintaining the cell's shape (Esus A. *et al* 2003).

The cytoskeleton is composed of three main components: the microtubules, the microfilaments, and the intermediate filaments. Microtubules are very dynamic structures, and in proliferating cells such as neuroblasts (neuron precursors), their probability of assembly is the same as that of depolymerization in all directions. This equilibrium results in the cell maintaining a spherelike morphology. However, during the differentiation of a neuroblast into a neuron, the microtubules become stabilized in specific directions, thereby generating the cytoplasmic extensions that will become the axon and the dendrites (Mitchison T and Kirschner M 1988).

It has been suggested that specific proteins may serve to stabilize microtubules and such proteins including the microtubule-associated proteins (or MAPs) MAP1A, MAP1B, MAP2, and tau. In support of this hypothesis, an asymmetric distribution of MAPs is seen in mature neurons, and tau is preferentially localized in axons (Binder LI *et al*., 1985). Furthermore, in pathological situations, tau has additionally been shown to be capable of forming aberrant fibrillar polymers.

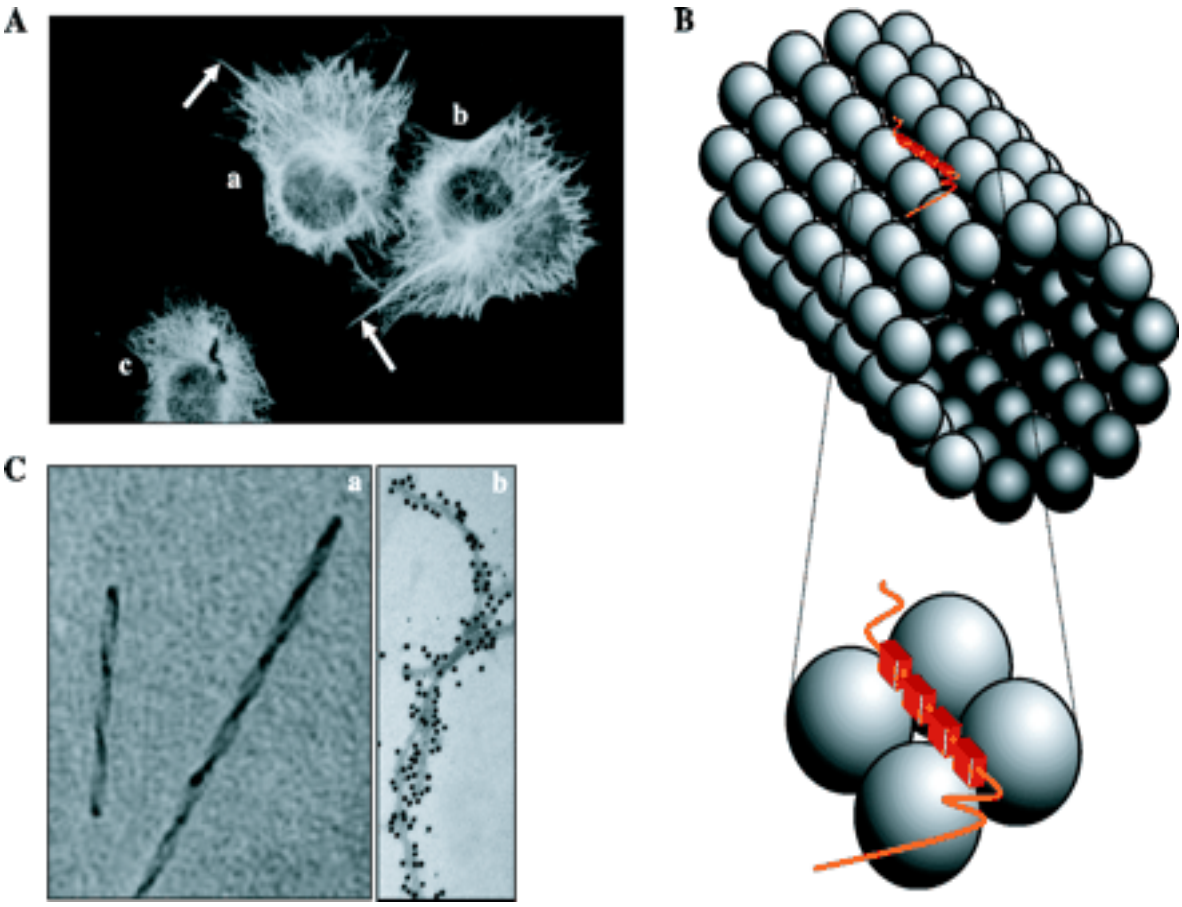


Figure 2.4: J. Avila - Tau is a microtubule-associated protein that stabilizes microtubules and that is able to self-aggregate in pathological conditions. A: the ectopic expression of tau in nonneural cells (a and b) promotes the stabilization of microtubules, leading to the formation of cytoplasmic extensions (arrows) that are not normally seen in those cells that are not expressing tau (c). B: scheme showing tau as a microtubule-associated protein (the size of the tau molecule is exaggerated compared with that of the microtubule). A view of the binding of tau to a tubulin dimer is indicated. C: in pathological situations (like Alzheimer's disease), the tau protein can form aberrant filaments (a). These filaments can bind to antibodies raised against the tau protein (b)

Tau proteins are microtubule-associated proteins that are found abundant in neurons in the central nervous system. Tau proteins were discovered in 1975 in Marc Kirschner's laboratory at Princeton University. Tau protein belongs to a group of proteins referred to as Microtubule-Associated Proteins (MAPs), that in common are heat resistant and limited affected by acid treatment without loss their function (D. W. Cleveland *et al.*, 1977).

The human tau gene is located over 100 kb on the long arm of chromosome 17 at band position 17q21 and contains 16 exons. Exon 1 is part of the promoter and is transcribed but not translated. Exons 1, 4, 5, 7, 9, 11, 12, and 13 are constitutive exons. Exons 2, 3, and 10 are alternatively spliced and manifesting in the adult brain. Exon 2 can appear alone, but exon 3 never appears independently of exon 2. In the central nervous system, alternative splicing of exons 2, 3, and 10 results in the appearance of six tau isoforms that are differentially expressed during development of the brain (N. Sergeant *et al.*, 2005)

Figure 2.5: Tau Protein Structure (Picture Courtesy from http://www.ebi.ac.uk/)

Tau proteins can interact with tubulin to stabilise microtubules and promote tubulin assembly into microtubules. There are two ways for Tau protein to control microtubule stability: isoforms and phosphorylation. Two different tau gene halotypes have been identified (H1 and H2) consisting of 2 common single nucleotide polymorphisms. H1 is the most common, and it is overexpressed in disorders like progressive supranuclear palsy and corticobasal degeneration.

In the brain tissue, there are six Tau isoforms that exist in the brain tissue, and are distinguished by their number of binding domains. There are three isoforms that have three binding domains and the other three have four binding domains. The binding domains are located in the carboxy-terminus of the protein and are positive-charged, allowing it to bind to the negative-charged microtubule. The isoforms with four binding domains are better at stabilising microtubules than those with three binding domains. The isoforms are a result of alternative splicing in exons 2, 3 and 10 of the Tau gene. Phosphorylation of Tau is regulated by a host of kinases, for example, Protein Kinase N1 (PKN), a serine/threonine kinase.

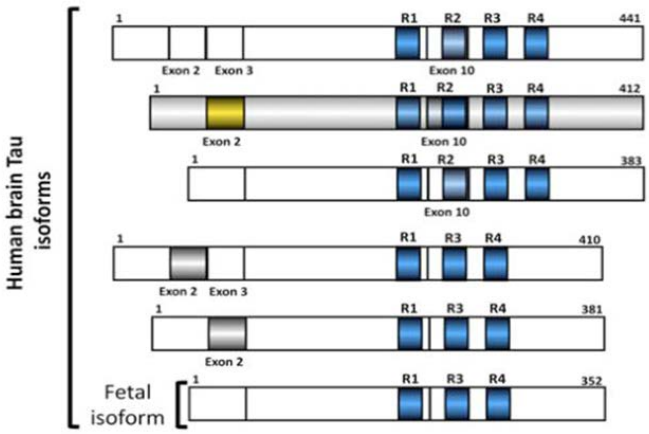


Figure 2.6: Six Major Tau Isoforms In Human Brain - Susanna Schraen

Tau Proteins can be used as biomarkers of brain pathology and as peripheral biomarkers of neurodegenerative diseases (S Schraen-Maschke *et al.*, 2008). Pathological tau may participate in the localization of Fyn kinase to the postsynaptic compartment, where it phosphorylates NMDAR subunits, causing increased inward Ca2+ conductance and leading to excitotoxity (S. M. Pritchard *et al.*, 2011).

Impaired interaction of Fyn kinase and hyperphosphorylated tau protein leads to hypomyelination and evolving demyelination of axons (C. Klein *et al.*, 2002). All these evidences indicate that the phosphorylated state of tau protein not only affects microtubule stability but also produces alterations on neuronal plasticity

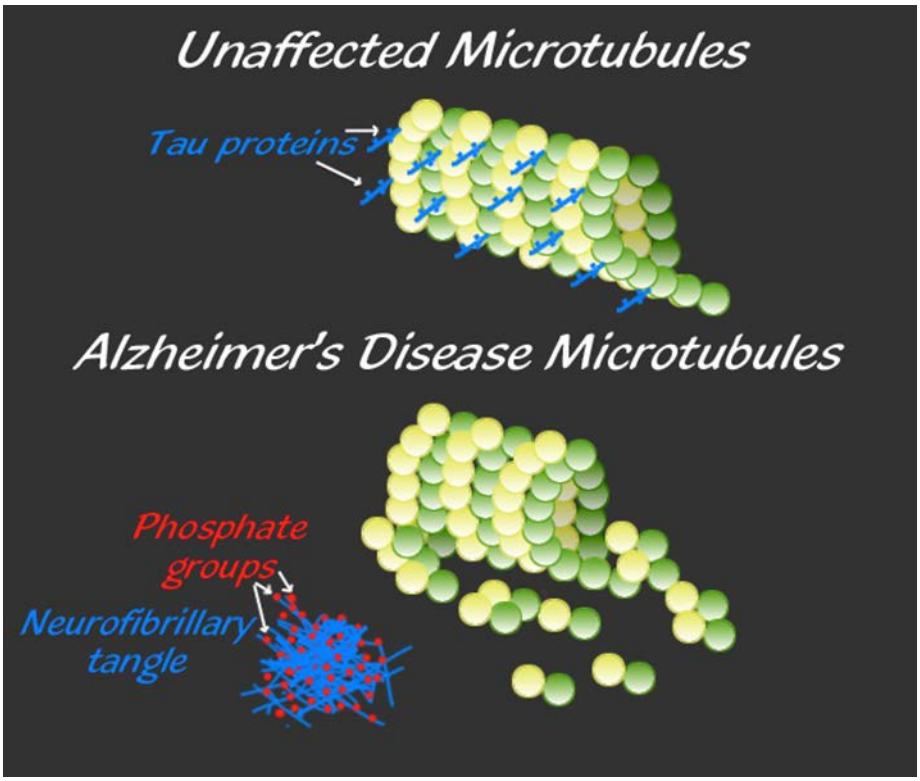


Figure 2.7: Structure Of Normal Tau Proteins And Alzheimer's Disease Microtubules With Neurofibrillary Tangle.

When tau proteins are defective, and no longer stabilize microtubules properly, they can result in dementias such as Alzheimer's disease. (Tan *et al.*, 2010) illustrated that increased cytokine levels after surgery leads to phosphorylation of tau protein and the formation of neurofibrillary tangles that associated with Alzheimer's disease. Whether these changes are serious enough to elicit neurodegeneration or cause a more subtle change in neuronal function remains to be determined

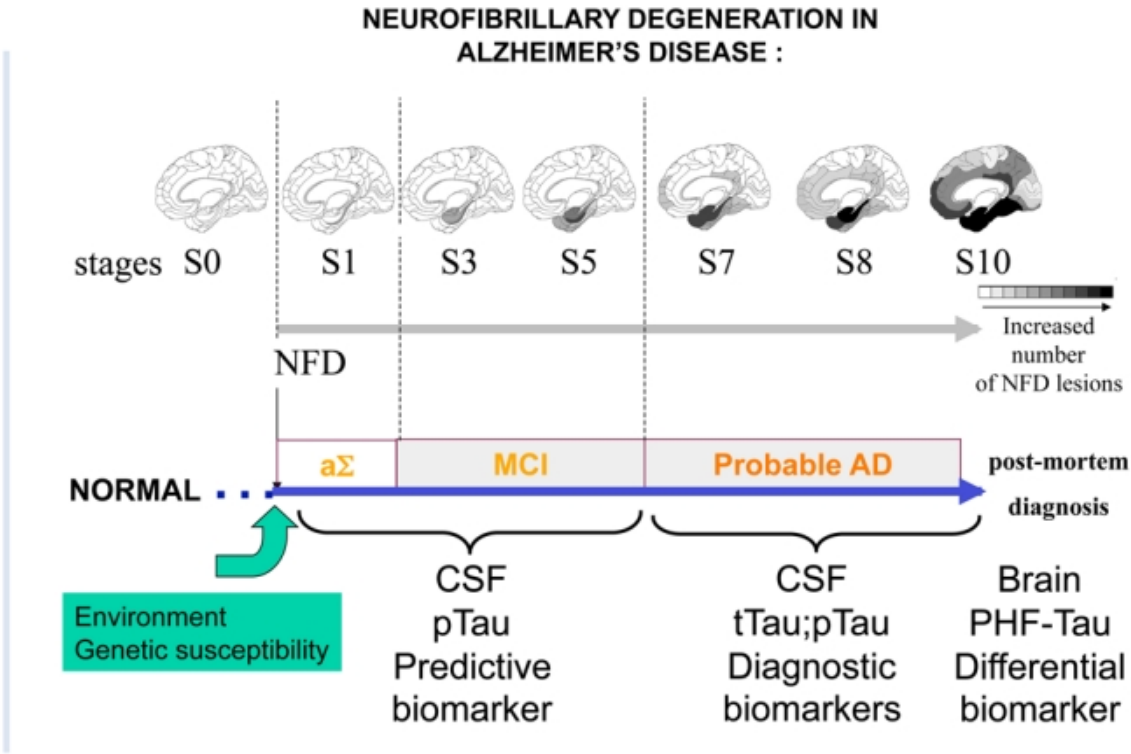


Figure 2.8: Stages In The Neuropathology Of Alzheimer's Disease. Implication For The Use Of Tau As Biomarker During The Course Of The Disease- Susanna Schraen

2.3 BRAIN DERIVED NEUROTROPIC FACTOR

Neurotrophins are an important class of signaling molecules in the brain responsible for axon targeting, neuron growth, maturation of synapses during development, and synaptic plasticity. This family of molecules includes nerve growth factor (NGF1) (Levi-Montalcini, 1966), brain-derived neurotrophic factor (BDNF) (Barde *et al*., 1982), as well as neurotrophins 3 and 4 (Hohn *et al*., 1990). Of these, BDNF is the best characterized in terms of its role in synaptic plasticity (Lohof *et al*., 1993; Levine *et al*., 1995, 1998; Kossel *et al*., 2001) and its potential role in the disease pathology or treatment of many psychiatric diseases (Duman and Monteggia 2006).

Brain derived neurotrophic factor (BDNF) is the most prevalent growth factor in the central nervous system (CNS). It is essential for the development of the CNS and for neuronal plasticity. Because BDNF plays a crucial role in development and plasticity of the brain, it is widely implicated in psychiatric diseases. The BDNF gene (in humans mapped to chromosome 11p) has four 5′ exons (exons I-IV) that are associated with distinct promoters, and one 3′ exon (exon V) that encodes the mature BDNF protein (Metsis *et al*., 1993).

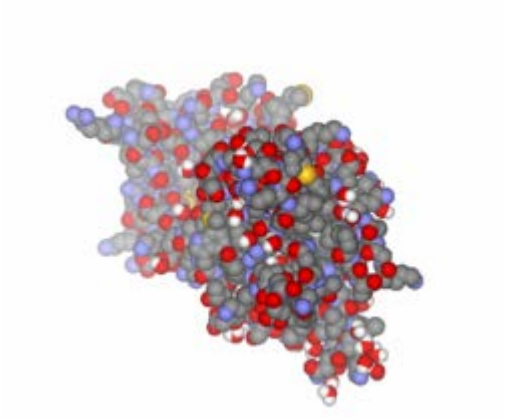


Figure 2.9: Brain-Derived Neurotrophic Factor (Wikipaedia 2013)

BDNF acts on certain neurons of the central nervous system and the peripheral nervous system, helping to support the survival of existing neurons, and encourage the growth and differentiation of new neurons and synapses. In the brain, it is active in the hippocampus, cortex, and basal forebrain areas vital to learning, memory, and higher thinking. BDNF itself is important for long-term memory. This gene may play a role in the regulation of stress response and in biology of mood disorders (Wikipaedia 2013).

BDNF is synthesized as a precursor protein known as prepro-BDNF that is cleaved into pro-BDNF, which can then be further cleaved into mature BDNF (Lessmann *et al*., 2003). Pro-BDNF and mature BDNF activate different intracellular signaling pathways (Woo *et al*., 2005; Matsumoto *et al*., 2008; Yang *et al*., 2009). Pro-BDNF signals through the low-affinity neurotrophin receptor p75 that is believed to be involved in apoptosis (Roux and Barker, 2002; Lessmann *et al*., 2003). Mature BDNF signals through its high-affinity tropomyosin-related kinase B (TrkB) receptor. When BDNF is bound to TrkB, it induces its dimerization and the receptor tyrosine kinase is autophosphorylated, leading to activation of intracellular signaling cascades, as well as augmentation of N-methyl-d-aspartate (NDMA) receptor currents (Levine *et al*., 1998).

There are at least three signaling transduction pathways that BDNF-TrkB activation can regulate. The phospholipase C γ (PLC γ) pathway, which leads to activation of protein kinase C; the phosphatidylinositol 3-kinase (PI3K) pathway, which activates serine/threonine kinase AKT; and the mitogen-activated protein kinase [MAPK, or extracellular signal related kinase (ERK)] pathway, which activates several downstream effectors. Each of these signaling pathways confer the unique function of BDNF on cells (Mattson *et al*., 2008).

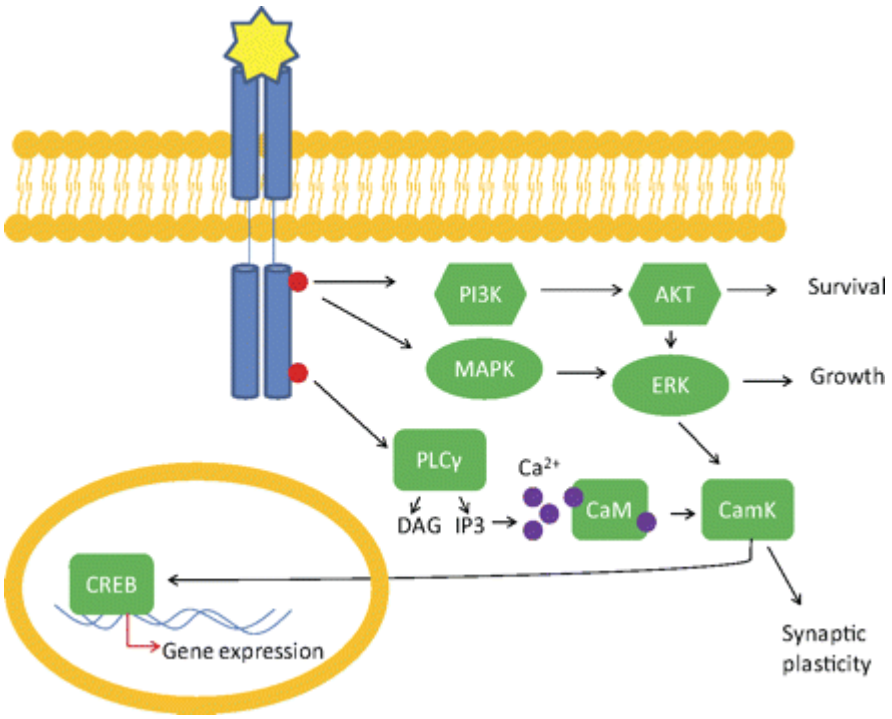


Figure 2.10: Overview Of BDNF Signaling Through Trkb Receptors

BDNF alone may not be sufficient to explain depression-related behaviors, but it remains an important risk factor for depression. Researchers have examined the role that BDNF plays in susceptibility to developing stress-related mood disorders, but preclinical investigations have not yet demonstrated how loss of BDNF alters vulnerability to stress (Advani *et al.*, 2009).

BDNF and NGF are important in the development, survival and maintenance of neurons in the central nervous system. The hippocampus has a key role in memory and spatial location since it is an important area of the brain required for learning and memory. Previous studies have demonstrated that the downregulation of BDNF and NGF in the brain may result in memory and learning deficits (Conner JM *et al.*, 2009).

Brain-derived neurotrophic factor number of studies has suggested that there is a strong link between volatile anesthetics, for example isoflurane, and cognitive impairment (Callaway Jk *et al.,* 2012, Lin D *et al.,* 2011). BDNF and NGF are important in the development, survival and maintenance of neurons in the central nervous system (Henriksson BG *et al.,* 1992). The hippocampus has a key role in memory and spatial location since it is an important area of the brain required for learning and memory. Previous studies have demonstrated that the down regulation of BDNF and NGF in the brain may result in memory and learning deficits (Conner JM *et al.,* 2009).

BDNF is involved in development, neurotransmission, and is also reactive to environmental stimuli it has been studied in the pathophysiology of Schizophrenia. It has been proposed that developmental abnormalities, as well as persistent cognitive and emotional dysfunction that occur in development, including circuit-level, genetic, or environmental alterations in BDNF expression, lead to pathological behavioral and neuronal features associated with schizophrenia (Durany and Thome, 2004). Post mortem studies of schizophrenia brain tissue have demonstrated alterations in BDNF in certain brain regions.

Various studies have shown possible links between BDNF and conditions such as depression, bipolar disorder, schizophrenia, obsessive-compulsive disorder, Alzheimer's disease, Huntington's disease, Rett syndrome, and dementia, as well as anorexia nervosa and bulimia nervosa (Zuccato C *et al.,* 2001).

2.4 POSTOPERATIVE COGNITIVE DISORDER

Post-operative cognitive dysfunction (POCD) is a severe complication characterized by cognitive decline in patients following anesthesia and surgery. Previous studies have suggested that volatile anesthetics, for example isoflurane, may contribute to such impairment (Zhang F. *et al.,* 2014). POCD may be self-limiting in the majority of patients; however, it may affect the prognosis and life quality of certain individuals (Steinmetz J, *et al.,* 2009).

Postoperative cognitive changes have been reported in elderly patients for over a century, and anesthesia has often been mentioned as a possible cause of this problem. In 1955, Bedford published a retrospective review of 1193 elderly patients who had surgery under general anesthesia during a 5-year period. He found that cognitive problems occurred in approximately 10% of older patients after surgery. Most of these patients experienced mild problems after surgery (inability to write a decent letter, concentrate, go shopping alone or read a book, increased forgetfulness after surgery, unable to attend to business), but were still able to function independently.

In a retrospective analysis of the International Study for Postoperative Cognitive Dysfunction (ISPOCD) research data, patients with postoperative delirium had a higher incidence of POCD one week postoperatively (Rudolph JL *et al.,* 2008). Although the ISPOCD results seem to suggest that postoperative delirium and POCD appear to be discrete events, other research provides support to the continuum hypothesis, specifically, that POCD is a subclinical from of delirium.

The exact pathophysiology of POCD remains undefined. Several mechanisms have been proposed to be involved in the development of cognitive impairments after surgery, including changes in cerebral blood flow, sleep disturbances, effects of anaesthetics, and inflammation. Hypoperfusion, hypoxia and the formation of micro-emboli have been shown to occur during and after surgery, and could potentially cause ischaemic brain damage, but a clear relationship with POCD has not been found (Krenk *et al.*, 2010).

The main risk factors for perioperative cognitive change are well recognized:

1. Non pharmacological factors:

- Increasing age
- Poor cognitive impairment
- Low educational level
- Depression

2. Intraoperative factors:

- Cardiac surgery
- Longer anaesthetic administration

3. Postoperative factors:

- Poorly controlled pain
- Postoperative wound infection
- Reoperation within 1 week of original operation

POCD is a subtle impairment of memory, concentration, and information processing that is distinct from delirium and dementia. Despite the fact that POCD is not a formal psychiatric diagnosis, the term is commonly used in the literature and is considered to be a mild neurocognitive disorder (Terri G. and Catherine C. 2011). The DSM-IV 2000 states that a mild neurocognitive disorder can only be diagnosed if the cognitive disturbance does not meet the criteria for three other conditions (delirium, dementia, or amnestic disorder). Post-operative cognitive disorders is different with postoperative delirium which tends to be a transient and fluctuating disturbance of consciousness that tends to occur shortly after surgery whereas post-operative cognitive disorders is a more persistent problem of a change in cognitive performance as assessed by neuropsychological tests (Newman *et al.*, 2007).

Postoperative cognitive dysfunction affects both young and old who present for surgery; however, the elderly have an increased incidence of this disorder due to less plasticity in the aged brain (Joseph W. Szokol *et al*., 2010). POCD (Postoperative cognitive dysfunction) can cause catastrophic loss of cognitive function, with associated increased mortality, risk of losing job & dependence of social welfare. (Steinmetz JC *et al.*, 2009).

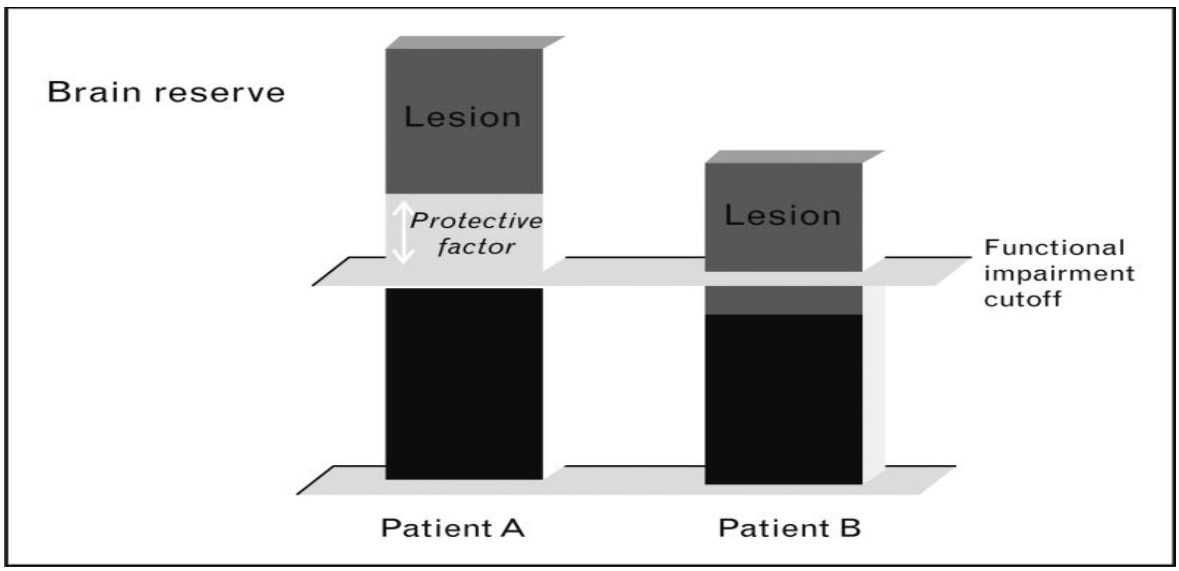
The clinical significance of POCD, particularly in older individual, is emphasised by evidence of the impact it confers on the ability to perform daily activities (Kojima Y, Narita M 2006). POCD is most common after cardiac surgery although up to 40% of people are affected for one week after non-cardiac procedures. Importantly, up to 15% of people continue to be affected after three months (Newman S *et al.*, 2007).

Several studies indicate that volatile anesthetics influence brain function and development beyond the time of anesthesia (Jevtovic-Todorovic V *et al.,* 2003). Further, clinical experience suggests that some patients have remnant symptoms after general anesthesia, including a broad range of neurocognitive dysfunction, though clinical studies have not found long-lasting effects of volatile anesthetics (Chen X *et al.*, 2001).

Volatile anesthetics affect cerebral bloodflow and metabolism (Lenz C *et al.*, 1998), and signaling via γ-aminobutyric acid receptors, N-methyl-d-aspartate receptors, and voltage-gated sodium channels (Ratnakumari L *et al.*, 2000). These effects occur during anesthesia, but not for later periods after withdrawal of anesthetic gases. These and other effects suggest that volatile anesthetics can have profound effects on cellular function, as well as altering expression at the message and protein levels (Kapinya KJ *et al.,* 2002). Sevoflurane induced relevant changes in protein expression profiles directly and 72 h after an anesthesia with 1 MAC (Kalenka *et al.,* 2007).

Several studies have found general anesthesia without surgery in old rodents induces prolonged changes in gene and protein expression and learning impairment that lasts days to weeks, implying either that the neurobiological machinery of memory is altered in an enduring way or that damage occurs (Culley DJ *et al.,* 2007). In addition, there is evidence that some volatile anesthetics promote processes implicated in the neuropathogenesis of Alzheimer's disease (Tang J *et al.,* 2009). Due to the subtle nature of POCD, it is often only the patient and/or partner who recognize the onset of this problem. The symptoms vary from mild memory loss to an inability to concentrate or process information.

The concept of cerebral cognitive reserve is often cited to explain why individuals with a similar degree of cerebral insult often have significant differences in the degree of cognitive symptoms. (Satz P *et al*., 1993) described a concept of brain reserve, which may help to explain vulnerability to postoperative cognitive problems: greater brain reserve serves as a protective factor, whereas less brain reserve serves as a vulnerability factor to a lesion or pathology.



In the figure 2.11: Patient A has greater reserve (which can be measured in a number of ways: education level, intelligence, brain size, white matter lesions, etc.) relative to patient B. Both patients experience a perioperative insult to the brain that results in a similarly sized 'lesion'. Patient A, however, because of cognitive reserve, does not demonstrate postoperative delirium or postoperative cognitive change and remains above the critical threshold for identifying a measurable change in functional outcome. Patient B with less preoperative cognitive reserve, by contrast, falls below this critical threshold and demonstrates significant changes in function which may manifest as postoperative delirium or POCD in the postoperative period.