NEUROPHYSIOLOGY

K.N. Sharma and S. Dua Sharma

Introduction

The history of modern Neurophysiology Research in India may perhaps be linked essentially to the pioneering works beginning from 1940s. Already the Status Reports of the work done in the country in the field and major achievements obtained upto 1980s or so has been covered (Desiraju, 1989; Sharma, 1994). The Present Section on Neurophysiological Sciences thus basically covers the last two decades with particular emphasis on current and emerging trends. It, however, shows some overlap with the earlier reports. This has been necessitated to maintain the continuity and it is in that spirit that a glimpse of the work done in earlier decades also finds its mention in the present Section.

The descriptions to follow are arranged in a manner that basic neurophysiological research coupled with applied and clinical applications are succinctly reflected with possible potential leads. As one will find, more and more emphasis at molecular and cellular level of investigation on the one hand and computational functions of the brain on the other are being pursued and form the emerging trends.

Neural Correlates of Feeding Behaviour

Following the pioneering work of Anand (Anand, 1961; Anand, 1963; Anand et al, 1964) identifying the dual hypothalamic feeding-satiety centers, lot of work has been done in the succeeding decades to indicate that these centers are influenced from higher limbic and other central neural regions and modulated by several factors such as state of nutrition, motivation and emotional factors, sensory qualities of food and hedonic matrix, as also the peripheral signalization from oral and gastrointestinal level (Sharma et al, 1977). The appraisal of food includes its taste, flavour, texture, viscosity, volume, temperature and other physicochemical characteristics (Sharma et al, 1975, 1979, 1995). At the post absorptive and systemic level, food as a nutrient acts as a metabolic signal on regulatory centres and through positive and negative feedbacks initiates modulation of the ingestive processes. Further, ontogenetic aspects of neural regulation of food intake have also been investigated in recent years (Sharma, 1975, 1992; Mathur et al, 1983, 1992) and provided the ontogeny calendar for various controls exerted on feeding behaviour and thereby on body weight of the adults. A series of papers were published by this group subscribing to the idea that most basic neural mechanisms regulating intake develop first and those for fine-tuning develop subsequently (Mathur et al 1992, 1986).

Sharma and co-workers have particularly contributed to the understanding of the role of impulses from gastrointestinal tract in feeding behaviour and showed

that the operation of modulating influences on alimentary receptors act at different levels of neuraxis (Sharma, 1992). The internal and external cues acting as sources of peripheral inputs bring about appropriate behavioural feeding responses through interaction with hypothalamus-limbic-sensorimoter cortical neuronal circuits (Kravtsov et al 1991, Sharma, 1992).

Sharma's group have also investigated the role of taste and other sensory qualities of diet, calorie intake and food deprivation (Radha Krishnan et al, 1986) and cross cultural biases in the regulation of feeding behaviour, food preferences and food habits (Moskowitz et al, 1975, Sharma et al, 1979). The differential effect of naloxone in intact and vagotomised rats suggests a possible involvement of endogenous opioid mechanisms in the gastro-gustatory interactions in taste (Radha Krishnan and Sharma, 1986). It is now an acceptable hypothesis that sensory activity of food and their consequent appraisal is a critical determinant of innate as also of acquired feeding responses ensuring an oral selection and metering of intakes.

It has been reported that gastric afferent discharge, analysed under varying conditions of nutrition and under various coupling effects of electrical, chemical and distension stimuli applied locally or at systemic levels, reveal distinguishing features, the modulating influence acting through a dual centrifugal control system, facilitation being routed through sympathetics, and inhibition through vagal fibres. The flow of information is not all in one direction. 'Tuning' of the receptors through the centrifugal controls via short and long feedback loops allows sensory pathways to act as variable filters so that stimuli tagged with a particular attribute or feature are alone allowed for detailed analysis (Sharma, 1992).

The design of a stereotaxic apparatus for frog brain was a major technical advance and helped in the precise localisation of neurons within specific regions of interest. Across- neuron pattern of the fasciculus solitarius and its gastric chemoceptive projections acting as a dipole field structure is an important contribution made from the Sharma laboratory during this period (Ramakrishna and Sharma, 1973).

Hedonic Matrix and Hypophagia: Singh, Selvamurthy and colleagues reported hypophagia on chronic intermittent exposure of albino rats to simulated hypobaric conditions. Subsequent investigations by Singh et al (1996, 1997a,) on high altitude hypoxic stress showed sensory dependence of feeding behaviour and changes in the hedonic responses, primarily in terms of increased palatability for sweetness, which could be due to anorexia-linked nutritional stress. Anorexia linked nutritional stress was reported to change hedonic responses in human volunteers exposed to altitude of 3500m (Singh et al, 1997b).

In recent years Pal and his co-workers demonstrated the important role of some key neurotransmitters in food and water intake (Pal et al, 2004).

Receptors and Cardiopulmonary Dynamics

The pioneering efforts in this country in the field of receptor physiology rightly belong to Prof.A.S.Paintal who significantly contributed to the identification and characterization of sensory receptors of the alimentary, cardiovascular and respiratory systems, in early fifties. Since then commendable contributions have been made by several groups in the country to further elucidate their functions and possible control mechanisms. Paintal's work on respiratory and cardiopulmonary afferent receptors has significantly advanced understanding in the fields of respiratory and cardiovascular medicine, pulmonary edema at high altitudes and other related problems (Paintal, 1973).

An important advance in J-receptor physiology took place in 1980 when Anand and Paintal (1980) showed that J receptors were stimulated by increase in pulmonary blood flow, and cardiac output. The reflex effects of J Receptors studied by Ravi (1988) showed that there existed a reflexogenic area in splanchnic bed that produced apnoea in vagotomised dogs. The most recent advances in J receptor physiology have come about following the observation that permeability increase during pulmonary oedema leads to a greater movement of the excitants across the capillaries leading to greater excitation (Anand et al, 1993).

With potential of considerable applied and clinical significance as reflected by the group led by Paintal, DST has opened and has been funding 'Exertional Breathlessness Studies Laboratory', at V.P.Chest Institute, Delhi to pursue these studies in terms of the leads obtained. Prof.Ashima Anand is heading the lab. Her current research interests have been to study the mechanisms that give rise to exertional breathlessness in various pathological states including CAD, obstructive diseases of the lungs as well as during high altitude pulmonary oedema (Anand A, 2005; Raj et al, 2005; Dehghani et al, 2004).

Neural Mechanisms of Sleep States

Studies conducted by H.N. Mallick and his colleagues at AIIMS have provided convincing evidence to show that the medial preoptic neurons form the most important part of the neural circuitry, which regulates sleep. A holistic approach, with simultaneous monitoring of sleep-wakefulness, thermoregulation and reproductive functions, have helped to establish the key role played by the preoptic area in the regulation of sleep-wakefulness. They studied the role of the noradrenergic afferent fibres, projecting to the preoptic area, in the regulation of several visceral functions, including sleep. This research group was the first to show the involvement of noradrenergic fibres in the induction of sleep. The group has also been a pioneer in demonstrating the restoration of functions after neural tissue transplantation. They were the first to show recovery of sleep after neural transplantation at the medial preoptic area. Electrophysiological recordings from the transplant were a significant scientific contribution. Magnetic resonance imaging of the lesioned preoptic area, done with the help of NMR was a rare achievement.

By employing fMRI, this group was the first to demonstrate the activation of the medial preoptic neurons during sleep. It was shown that the medial preoptic area has the homeostatic mechanism for fine-tuning food intake, body temperature, locomotor activity and sleep-wakefulness (Kumar, 2003; Ray et al, 2004; Khubchandani et al, 2005; Srividya et al, 2005; Vetrivelan et al, 2005; Vetrivelan et al, 2006a; Vetrivelan et al, 2006b; Srividya et al, 2006;).

REM Sleep

B.N.Mallick and his colleagues working at JNU have added another significant dimension to the understanding of sleep states, not known earlier. His group has pioneered the studies particularly on REM sleep. The loss and / or disturbances in this stage of sleep are associated with several somatic, psychic, developmental and other disorders but the mechanism of its regulation, function and mode of action have remained largely unknown and form the major thrust of Mallick's work, who used from behavioural to molecular and cellular level models.

In freely moving normally behaving cats it was shown for the first time by simultaneous recording of a pair of REM-OFF and REM-ON neurons that temporarily they go off and on, respectively, in relation to spontaneous REM sleep and they behaved identically during stimulation induced wakefulness (Mallick and Joseph, 1998). Working on rats, the group showed convincingly that the cholinergic inputs from the REM-ON neurons excite the GABA-ergic neurons, which in turn inhibit the REM-OFF neurons for the generation of REM sleep (Mallick et al, 2001, Kaur et al, 2001). This was a significant advancement in the knowledge for understanding the neurochemcial regulation of sleep. Later studies confirmed the role of interaction of adrenergic and GABA-ergic inputs in the pedunculo-pontine tegmental area REM-ON neurons on the regulation of REM-ON neurons and thus on the regulation of REM sleep (Pal and Mallick, 2006).Further studies led to the conclusion that the area in the brain which induces wakefulness excited the REM-OFF neurons and inhibited the REM-ON neurons (Thankachan et al, 2001), while the area in the brain which induces sleep had opposite responses (Mallick et al, 2004a). It was also observed that REM sleep loss was associated with decrease in body temperature (Mallick et al, 2004b) and that REM sleep prevents Adoptosis (Majumdar and Mallick, 2005).

Neurophysiological Co-relates of Nociception and Pain

The Pain Laboratory at AIIMS, headed by Prof.R.Mathur is primarily exploring in recent years the neural mechanisms involved in pain perception and its alleviation with the use of sucrose therapy. Experiments are being conducted using rat as an animal model to test the hypothesis that sucrose therapy may alleviate chronic pain. Studies are ongoing using human subjects in collaboration with the Department of Neurology.

The group has provided sufficient comparative data to demonstrate involvement of hippocampus, cingulate gyrus, amygdala, septum and hypothalamus in the modulation of pain. They were the first to demonstrate that each area has its special unique feature of the control. For example activation of ventromedial hypothalamus reduces pain via its glucoresponsive neurons in relation to the food such as ingestion of palatable food. Palatable food ingestion relieves moderately chronic severe repetitive stress of unpredictable type interval and intensity.

The group designed a test namely sucrose challenge test utilizing the above-mentioned observations to determine the pain status of an individual. It is cheap, economical, safe, quick and a sensitive tool in the hands of a scientist as well as clinician. It has been utilized to: i) differentiate early cases of Alzheimer Disease patients from dementia of other causes; ii) demonstrate greater efficacy of yogic life style intervention than use of NSAIDS or BOTOX in chronic tension type headache, and iii) demonstrate efficacy of ganglion block in Trigeminal neuralgia patients (Jain et al, 1999; Jain et al, 2000a Mukherjee et al, 2000; Dutta et al, 2001; Mukherjee et al, 2001a; Mukherjee et al, 2001b; Mukherjee et al, 2002).

They have also reported the mechanism underlying attenuation of pain, relief of stress and deficit in memory produced by intermittent chronic exposure to magnetic fields. Recent research effort is to examine motor responses to noxious/ non-noxious stimuli in spinalised humans and rats and find out the role of electromagnetic stimulation in recovery. Spinal injury de-links the motor cortex, spinal central pattern generators (CPGs) and peripheral receptors. Besides, it also provides a hostile biochemical and cellular micro-environment for recovery in terms of biochemical and cellular profiles. Studies are also being conducted to find out the pattern of recovery in both the sensory and motor functions after embryonic stem cells transplantation locally, magnetic stimulation of motor cortex and spinal cord and peripheral sensory stimulation with a view to re-establish the loop of information flow from motor cortex to spinal cord, periphery to spinal cord and vice-versa (Mathur et al, 2008).

The group led by Gupta and Goyal at PGI, Chandigarh has been working on the newer perspectives of pain pathways and elucidated the role of neuronal marker; proto-oncogene c-fos (Gupta and Goyal, 2005). It has been shown to be an invaluable tool in the study of pain as it can be used as specific marker to map out regions corresponding to noxious stimulation (Muglani and Hunt, 1995) and the effects of analgesic or anaesthetic agents in the treatment or prevention of pain (Sun et al, 1996). Studies by Mediretta and colleagues at UCMS have shown that oxytocin exhibits an antinociceptive effect in formalin test and tailflick test in mice and suggested the involvement of kappa – and delta – opioid receptors in the response (Mediretta et, 2005). Sharma and others also from the same group in UCMS, highlighted the role of COX-2 rather than COX-1 isoenzymes in mediating antinoceceptive effects and their role in opioid tolerance / dependence (Sharma et al, 2005).

Studies conducted by Bhattacharya and her group also working at UCMS have further revealed the cortico-brainstem interaction in modulation of both nociceptive and antinociceptive responses (Bhattacharya and Sharma, 1986, Bhattacharya et al, 1988). The group also showed involvement of some new brain indices in acupuncture pain treatment effects (Bhattacharya et al, 1991). A comparison of autonomic functions of post herpetic neuralgia and herpeszoster patients with normal controls showed decreased vagal / increased sympathetic activity in comparison to HZ and normal controls (Bhattacharya, 2005).

Kanaka reported that nociceptors activated by algesic chemical substances can modulate the profile of non-nociceptive afferents like deep pressure (Kanaka, 1988). The possibility of such interaction and inhibitory modulation being peripheral in locus was convincingly put forward (Kanaka et al, 1989).

Sujatha from MSRDC, Bangalore brought out clear dangers posed to oral health from smoking and chewing tobacco, particularly as applied to Indian population and linked to their life style habits such as smoking, betel chewing and excessive alcohol consumption. Pain was rarely found to be an early symptom and therefore it was pointed out that all tobacco users need regular dental checkup so as to reduce overall morbidity and mortality from oral cancer and other mouth disorders (Sujatha, 2005).

Venkatesh and his co-workers in MSRMC, Bangalore have been working to see the influence of psychological stress on pain perception, sensory motor integration and central processing speed. They have demonstrated the analgesic potential of sex hormones and psychological stress in animal models, and established the impact of psychological stress on audio-visual response latency in human volunteers (Venaktesh et al, 2002; Sudhakar and Venkatesh, 2003a; 2003b).

Psychoneuroimmune Responses and Evoked Potentials

In a series of studies undertaken at UCMS, Delhi, Tandon and his colleagues have shown that neural substrate in medial basal hypothalamus (MBH) involving LHR, catecholamines and Beta-endorphins, regulates not only endocrine events but also those involved in immune responses. These studies later led to understanding and subsequent development of GnRh analogues used in control of human reproduction (Kreig et al, 1976; Tandon et al, 1983; Tandon and Sharma, 1986; Tandon, 1992).

In the field of evoked potentials studies (EPs), Tandon established normative data for North Indians (Yadav et al, 2002), for brainstem auditory evoked responses, visual evoked potentials and P300 responses, and in women in normal pregnancy (Tandon et al, 1990) and menopause (Khaliq et al, 2005), and also showed the importance of EPs in evaluating derangement in sensory functions in states of protein calorie malnutrition (Vandana and Tandon, 2006), clinical cases of

xerophthalmia (Tandon and Ram, 1993), and cognitive dysfunctions in chronic pain patients (Tandon et al, 1997). These studies suggest that nutritional insult to brain in its critical period of growth and development (prenatally) adversely affects the functional integrity of auditory pathways in the brain stem of the new born. His group's more recent work on environmental pollutants in factory workers has shown adverse influence on brain sensorium and include impaired cognition (Kumar and Tandon, 1997). Primary hypertension affects sensory and cognitive functions of the brain (Ram et al, 1996) and so does diabetes (Tandon et al, 1999). Also B12 and iron deficient children show cognitive decline which may be reversed by nutritional supplements (Shankar et al, 2000).

Roopkala and her co-workers at MSRMC recorded compound muscle action potentials (CAP) bilaterally from alnae nasi muscles following stimulation of facial nerve in healthy adults. Latency, amptitude and total duration of the CAP were compared on both sides. A significant positive co-relation between the 3 parameters with the highest for the amplitude was observed, suggesting amplitude as a better parameter for comparison while testing for side to side facial nerve integrity (Roopkala et al, 2003). Somatosensory evoked potential (SEPs) after median nerve stimulation in the wrist of totally blind and normal sighted persons were also studied. It was observed that congenitally blind individuals have larger N20 amplitude, suggestive of greater Somatosensory Cortical representation (Dayanand et al, 2008). Elizabeth and her colleagues have been working in the field of psychoneuro-immunology and observed that leptin the adipocyte hormone acts as a neuro-immunomodulator in academic stress and also as a satiety factor in androgen replacement studies in hypogonads (Elizabeth et al, 2008). Ambarish and his co-workers have been studying the behaviour of plasma pro-inflammatory cytokines with bouts of exercise versus regular moderate exercise in MBBS students, practitioners of yoga, patients of Rheumatoid arthritis and patients of diabetes mellitus. It has been observed that the plasma levels of pro-inflamatory cytokines (IL-6 and TNF-L) fell below baseline levels after regular moderate exercise of one month indicating that such individuals are less prone to inflammation (Ambarish et al, 2005). Taking a lead from present observations, efforts are now being made to further examine the possible neurohumoral mechanisms underlying these cytoimmunological profiles. Bhaskar observed that L-Dopa therapy in patients with Parkinson's disease caused recovery of P100 latency of visual evoked potentials. Pattern stimulus evoked potentials were not improved. (Bhaskar, 1986).

Brain Functions at High Altitude: DRDO Perspective

Neurophysiological changes under stressful environmental conditions has been an area of thrust in Defense Research and Development Organisation (DRDO) as troops in India are posted to different environmental situations like high altitude, hot and cold environment, noise and vibration in naval dockyard and ground personnel in airfield. All are exposed to varied kind of environmental stresses. In this context the contribution of defense neuroscientists has been quiet substantial.

Research on brain functions at high altitude started in India way back in 1970s pioneered by Dr. W. Selvamurthy at Defense Institute of Physiology and Allied Sciences (DIPAS), Delhi. The 70s and 80s saw extensive research on effect of high altitude exposure on cognitive functions of volunteers. Initial studies focused on neuropsychological alterations, responses of autonomic nervous system, electroencephalogram, sleep profile, hypothalamic thermoregulatory efficiency and effects of sleep deprivation on high altitude exposure.

In 1990s much of the focus on neurophysiological research at high altitude was on evoked potentials, EEG patterns and sleep disturbances, event related potentials and hedonic matrix. Psychological and electrophysiological tests for evaluation of cognitive functions of human volunteers inducted to high altitude were carried out that provided relevant information regarding various aspects of functioning of the brain following high altitude exposure.

The early phase of the present decade has seen a renaissance in neuroscience research with the thrust area being the neurotransmitter and molecular mechanisms responsible for the altered neurophysiological functions responses observed at high altitude. In addition to human studies, investigations have been carried out on animal models to understand the molecular events pertaining to hypoxic stress. In addition to the studies conducted at DIPAS, the basic mechanisms pertaining to hypobaric hypoxia induced memory impairment were also studied at National Institute of Mental Health and Allied Neurosciences, Bangalore (NIMHANS) and National Centre for Biological Sciences, Banglore (NCBS).

Autonomic Response: Series of studies were conducted by Selvamurthy and colleagues to evaluate the neurophysiological responses in young healthy soldiers during acclimatization at 3,500m altitude in Western Himalayas. The responses of autonomic nervous system was studied in fresh inductees during three to five weeks of acclimatization at high altitude and compared with those of one year acclimatized lowlanders and high altitude natives (Selvamurthy 1988). Parasympathetic predominance was observed in high altitude natives while a relative hyperactivity of sympathetic system was noted in sojourners on immediate arrival to high altitude that gradually recovered following stay for a week due to onset of acclimatization. The relative parasympathetic dominance observed in high altitude natives showed a gradual decrease during their sojourn on the plains.

Yoga and Brain Function: In addition to basic research on the electrophysiological responses on high altitude exposure, studies were also carried out to evaluate the therapeutic efficacy of yogic exercises in ameliorating stress induced alterations in neurophysiology. Studies by Selvamurthy et al (1998) on effect of yogic exercises on progressive autonomic readjustments showed gradual improvement in baroreflex sensitivity and changes in alpha index of EEG, indicating progressive attenuation of sympatho-adrenal and rennin-angiotensin activity. Similar

studies by Ray et al (2001) on effect of yogic exercises on physical and mental health of young trainees showed improved psychological functions along with decreased sympathetic activity on training for 5 and 10 months.

EEG and Sleep Disturbances: Study on changes in EEG pattern during acclimatization to high altitude by Selvamurthy et al (1978) revealed cortical depression in the initial phase of induction which changed to cortical desynchronization in the later part of the first week of induction as a result of sympathetic hyperactivity while the cortical neurons gradually adapted to lower Pa CO_2. During acclimatization there was a gradual build up in EEG waves which was observed both in acclimatized individuals and high altitude natives. Alterations in sleep pattern with changes in EEG patterns and frequent arousals, awakening and reduction in slow wave sleep were observed on high altitude induction that persisted till second week of stay at high altitude (Selvamurthy et al 1986). This is an adaptive feature to prevent the accentuation of hypoxemia which is known to occur during sleep hypoventilation since individuals lacking such responses are likely to suffer from acute mountain sickness. Sleep deprivation studies conducted by Selvamurthy et al (1986) reported augmented sympathetic reactivity to an additional stress as evidenced from the increased cold pressor response following sleep deprivation.

fMRI and Meditation: Studies carried out by Khushu et al (2000, 2005) on the functional mapping of the brain using fMRI during meditation suggest brain activation pattern which are related to meditation experience and dedication. Studies suggest better understanding of the effects of meditation will also help in considering possible applications of this practice, for improvement of mental health and a concomitant reduction in stress level. This is especially relevant to personnel exposed to stressful situations. The regular practice of meditation results in training of these neural networks associated with executive functions particularly the attention.

Evoked Potentials: Delayed sensory conduction has also been reported by Mukhopadhyay et al (2000) by analyzing auditory evoked potentials in human subjects inducted to an altitude of 3500 m. Similar delay in wave I and wave III of Auditory Brainstem Responses was observed in altitude dependent studies . While the N1, N2 and P2 latencies were not significantly affected at 3200m and 4300m, there was a significant increase in latency of P3 component of Auditory Event Related Potential in human subjects indicating slow signal processing (Singh et al 2004a). Visual Evoked Potentials on the other hand showed increase in N1wave latency at 4300m (Singh et al 2004b).

Cognitive Functions: Decline in overall psychomotor efficiency, accuracy and speed was registered during early stage of exposure to an altitude of 4000 m. Sharma et al (1976) reported change in concentration in male subjects following exposure to 4000m. An interesting study by the group showed better psychological

performance and toleration to altitude stress by Gorkha in comparison to Rajputs and Madrasis. They have carried out recording of cognitive potentials in actual field conditions on soldiers posted at HA. The altitude and time dependent changes in Event Related Brain Potentials recorded showed alteration in P3 wave latency indicating slow processing of stimulus evaluation following high altitude exposure (Singh et al 2003).

Titus et.al. (2007) at NIMHANS and Maiti et. al. (2007) at DIPAS have shown denitrific atrophy and neuronal degeneration in several brain regions including the hippocampus, cortex and striatum. Hota et al. (2007, 2008a, 2008b), have reported memory impairment and occurrence of glutamate excitotoxicity while Barhwal et. al. (2007, 2008) have demonstrated oxidative stress, mitochondrial dysfunction and apoptosis on simulated high altitude exposure.

The role of neurotrophins and p75 NTR in mediating neuronal degeneration in hypobaric hypoxia has also been studied by Hota et. al.(2008). A significant contribution in this regard is the elucidation of NMDAR mediated regulation of p75 NTR expression. The therapeutic efficacy of N-acetyl cysteine (Jayalakshmi et. al. 2005, 2007), acetyl L carnitine (Barhwal et. al. 2008) and Bacosides (Hota et. al. 2008) has also been evaluated under hypoxic stress.

Though several milestones have been achieved in pursuit of understanding the brain functions at high altitude, the precise molecular mechanisms and interplay of neurotransmitters involved under stressful conditions still remains quiet elusive. This greatly limits the use of therapeutic interventions to ameliorate stress induced alterations in brain functions. However, with the present pace of advancement in research on brain functions under stressful conditions, neuroscientists in India are sure to leave a mark in the international arena.

Neurophysiology and Yoga

Inspite of long history and investigations on yoga in India (Kuvalayananda, 1925; Anand et al 1961; Wenger et al 1961), much interest has been aroused in recent decades. Patel and North (1975) showed the usefulness of yoga in management of psychosomatic conditions. Extensive studies at Swami Vivekananda Yoga Research Foundation (SVYASA), Bangalore, have shown the usefulness of Pranayama in conditions like bronchial asthma (Nagarathna and Nagendra, 1985). Datey and his colleagues (Datey et al, 1969) showed the useful role of Shivasana in managing hypertension. Studies at AIIMS, Delhi have shown the effectiveness of life-style change for various psychosomatic ailments (Bijlani et al, 2005).

How well we are familiar that yoga practices intermesh with the nervous system at several levels, ranging from control of postures to associate / disassociate with different sensory modalities, enter subtle planes to impart mastery of mind, will and drive and transcend into realms not virtually understood. For instance,

one consistent textual and experiential fact is the attenuation of autonomic tone by means of pranayama, perceptual control of sensory awareness by consistent practice, and the existence of a close link between oropharyngeal sensory and motor activity and inspiration both in deglutition and phonation of mantra, japa or chanting, a feature that may have some basis in high muscle spindle density in the facial muscles.

In early 1980s another important step in yoga research was taken by the Indian Council of Medical Research which funded a multi-faceted project to understand the neurophysiological changes in *pranayamas* and meditations under T. Desiraju at NIMHANS, Bangalore (Desiraju, 1983). The project used what were, at the time, state-of-the-art assessment techniques including different modalities of evoked potentials, fast-Fourier transform analysis of the electroencephalogram, polysomnography, and various methods to assess neurotransmitter levels and their metabolites. The main research methodology innovation was that subjects were studied in repeat sessions to assess intra-individual variability and each subject was assessed in 'experimental' and 'control' sessions (i.e., a self-as-control design, rather than a matched groups design, which was used in most studies before this (Telles and Desiraju, 1993; Telles et al, 1993).

It was also considered interesting to understand whether yoga would be of use in other applications, such as stress management, or in maintaining homeostatic functioning under adverse environmental conditions. Research at DIPAS, Delhi showed that yoga practice helps soldiers adapt to extremely difficult and demanding environmental conditions (Rawal et al, 1994; Selvamurthy et al, 1988). Very recently, a study by SVYASA, Bangalore demonstrated that yoga can even help in extreme environmental adversity, based on a post-tsunami study conducted in the Andaman Islands (Telles et al, 2007).

While various centers across the country are continuing to study therapeutic and other applications of yoga, most researchers have more recently attempted to understand *how* the benefits are obtained. Most relevant perhaps are studies on depression (Janakiramaiah et al, 2000), schizophrenia (Duraiswamy et al, 2007), epilepsy (Sathyaprabha et al, 2008), irritable bowel syndrome by Deepak at A.I.I.M.S., Delhi (Taneja et al, 2004), and studies on yoga for complicated pregnancies (Narendran et al, 2005) and on discomfort related to perimenopause, respectively, at SVYASA, Bangalore (Chattha et al, 2008).

There have also been attempts to specifically understand changes in the nervous system related to yoga practice, using evoked potentials (Panjwani et al, 2000; Raghuraj and Telles, 2004), transcranial Doppler (Naveen et al, 1999), functional magnetic resonance imaging (Naveen, and Telles, 2003), and polysomnography (Sulekha et al, 2006).

Yoga lab at UCMS

The yoga lab at UCMS under the leadership of Prof.Savita Singh is at the forefront of research as far as the role of yoga visa vis possible neurophysiological co-relates in modern day medicine are concerned. The work mainly covers the role of yoga in modern day lifestyle diseases like diabetes mellitus, coronary artery disease and hypertension. In a study on NIDDM patients, plasma fasting glucose and 1 hr post prandial glucose levels were found to be significantly reduced on performing Yogasanas (Malhotra et al, 2005). In another study, the B.P. was also found to be significantly decreased in DM patients on yogic practice (Singh et al, 2004c, 2004d). Yogasanas have also found to improve nerve conduction in mild to moderate type II DM patients (Malhotra et al, 2002). In a recent study, yoga improved the latency of P300 (a marker of cognition) and visual evoked potentials in DM patients (Kyizom, 2007). Study conducted on coronary artery disease patients has shown a decrease in sympathetic activity and increase in parasympathetic activity on doing yoga (Rajkumari, 2006).

Along with the yoga lab, the neurophysiology lab has become one of the important centers for research on humans using evoked potentials. Various evoked potentials like auditory brainstem response (ABR), mid latency response (MLR), slow vertex response (SVR), visual evoked potentials (VEP) and P300 are used for research as well as diagnostic purposes. The lab has reported changes in the above mentioned parameters in various states like hypothyroidism, iron deficiency anemia, dementia etc (Vaney et al, 2002; Bandhu et al, 2003; Anjana et al, 2008). In conjunction with the yoga lab the group is also evaluating the effect of yoga on the brain using the above mentioned EP tests.

Jaisri and her group has undertaken a number of projects in collaboration with the Department of Yogavidya Pranic Healing in the M.S.Ramaiah Medical College and Hospital. The basis of the studies is the presence of a Neuro-immuno-psychological and neuro-immuno-endocrine axis that helps in the process of self-recovery and thus provides relief from most of the symptoms and contributes to the improvement in lifestyle. Some of the studies done by the group include the Effect of Yogavidya Pranic Healing on the control of blood pressure, coronary artery disease, improvement in lung function tests, improvement in quality of life and so on. Event Related Potential, evoked potentials and electroencephalogram have been used for the study (Jaisri et al, 2007; Vrunda et al, 2004). The results suggest increase in theta and alfa activity of the brain and decrease in beta activity during meditation.

Applied Neurophysiology

The group at NIMHANS under the leadership of Bindu M Kutty has been studying the age associated changes in normal healthy subjects with practitioners of yoga and demonstrated that yogic practices prevented the age associated alterations in sleep structure, quality and EEG Power Spectra (Sathiama et al,

2006). The group has also delineated the role of subiculum in consolidation of spatial memory and interface in processing and transferring the spatial information from hippocampus and cortex (Bindu et al, 2007). Laxmi's group has been working on neural networking activities in the amygdalohippocampal circuits during various stages of sleep and showed clear alterations in sleep morphology on exposure to acute immobilization stress (Preethi et al, 2008).

TR Raju's group has suggested that chronic stress produces consistent and reversible changes within dendritic arbors of CA 3 hippocampal neurons, characterized by decreased dendritic length and reduced branch number. His research has also demonstrated that entorhinal cortex lesion could prevent the stress-induced damage in CA3 hippocampal neurons and thus explain the excitotoxicity mechansisms in the hippocampal atrophy due to stress (Sunanda et al, 1997). Group led by Shankar Narayan Rao emphasized that restoration of hippocampal atrophy to stress can be carried out with an enrichment (Dhanush Kodi et al, 2007; Ramkumar et al, 2008).

Another area of research has been to understand the pathology of Amyotrophic lateral selerosis (ALS), undertaken in in-vitro and in-vivo rat models developed in the lab using CSF from ALS patients. Using these models the molecular mechanisms involved in degeneration of motor neurons are being pursued. Therapeutic effects of various trophic factors including BDNF, CNTF, VEGF and several other compounds such as deprenyl and cyclophosphamide have been investigated (Shobha et al, 2007; Senthil Kumar et al, 2007). Recently Subramanian from IIT, Kanpur has shown how SODI protein misfolding / aggregation can cause the motor neuron disease using molecular modeling and cell culture systems.

Among other neurodegenerative conditions Parkinson's disease (PD) has attracted a lot of attention. In India there are several research groups focusing their work on different aspects of this disease. Among them, Vijayalakshmi Ravindranath, Director, National Brain Research Centre (NBRC), Manesar, Haryana has been interested in understanding the molecular mechanisms involved in this selective vulnerability of Parkinson's disease for almost two decades. Her laboratory had earlier shown that redox perturbation of protein thiols underlies the mitochondrial complex I dysfunction, and seen in animal model for Parkinson's disease. It is their hypothesis that thiol modification occurring as a result of oxidative stress results in mitochondrial dysfunction and altered downstream redox signaling leading to activation of cell death pathways in a region specific manner. They are investigating the early events in terms of activation of the cell death pathway and the suppression of the cell survival pathways. The incidence of PD is lower in women and estrogen is a potent neuroprotector. They also investigate the mechanism underlying the neuroprotection afforded by estrogen with a goal towards developing neuroprotective strategies for PD.

Similar research is also being carried out by Mohanakumar and his group at Indian Institute of Chemical biology, Kolkata and focuses his research on identifying the molecular basis of neuronal loss in Parkinson's disease, motor neuronal disorders and Huntington's disease employing animal models, human postmortem brain tissues, primary cell cultures, cell lines and modified cell lines (cybrids). The approach involves delivery of toxins and drugs to discrete neuroanatomical loci in animals, analyses of behavioral abnormalities, neurotransmitter metabolism, reactive oxygen species, cellular events of apoptosis, membrane potential/currents, mitochondrial functions and mitochondrial gene expression. Antiparkinsonian drug screening is a major activity.

Neurophysiology based neurorehabilitation techniques as applied to humans is a unique approach, undertaken by Dr.Vaijayantee Belle and her group at Pune. It applies concepts of dynamic system theory and cybernetics to the functioning of the nervous system. It also involves tools and methods for objective documentation of re-organization of nervous system. The co-ordination Dynamics Therapy (CDT), a movement-based learning therapy, has been used and shown to be effective in significantly improving the functioning of the injured Central nervous system in a variety of conditions (Schalow et al, 2008).

Brain Computational Functions and Cellular Matrix

The group led by Dr.Bhalla and his co-workers at NCBS, Bangalore, has substantially contributed to several emerging trends. His group has mainly focused its attention on the computational function of the brain, and how it arises from the elementary properties of biochemical and biophysical systems. They have used olfactory processing and memory as two revealing systems from which to launch their exploration of these issues.

At the macro level they used parallel electrical recordings to study detection and signal encoding (receptors, olfactory bulb) right through to the storage of olfactory memories (piriform cortex, hippocampus). They have been developing techniques to read out the connection matrices of these circuits at larger scales and higher resolutions, to better understand network computation. Animals are trained on odor memory tasks to put these recordings into a functional context. At the micro level, they use microscopy and detailed computer simulations to explore the networks of signaling chemicals that operate in neurons. A key finding of their work is that inside cells, beyond the slow "calculations" of DNA, the signaling system itself operates as an independent computing system that can process and remember information. They have identified families of pattern-detectors and memory 'switches' and are beginning to look at how these work in the extended spatial context of the cell. They believe that this convergence of experiments and models across scales is a good way to gain interesting insights into the function of big and small networks in the brain. (Bhalla and Iyengar, 1999; Le Novere et al,

2005; Hayer and Bhalla, 2005; Rajan et al, 2006; Ajay and Bhalla, 2007; Bhalla, 2008; Ramakrishnan and Bhalla, 2008; Khan et al, 2008).

Dr. S.Chattarji of Dr.Bhalla's group has been studying the effects of stressful experiences on synapses, cells and microcircuits in the hippocampus and amygdala, by using a combination of behavioral, neuroanatomical, computational, genetic engineering and electrophysiological techniques. Using this strategy, they have identified several novel neural correlates of stress-induced plasticity in the amygdala, which are strikingly different from those observed in the hippocampus. The findings suggest that prolonged stress leaves its mark by enhancing both the physiological and structural basis of synaptic connectivity in the amygdala, thereby triggering the emotional symptoms observed in stress-related psychiatric disorders (Vyas et al, 2002, 2003; Narayanan et al, 2005; McEwen and Chatterjee, 2006).

The group led by Dr. Hasan addresses the question of how a cell responds to changes in levels of calcium within it. They are specifically interested in the second messenger Inositol 1,4,5-trisphosphate (InsP3) and its intracellular receptor - the InsP3 receptor. This protein exists on the membranes of intracellular calcium stores and performs the dual function of a receptor for InsP3 and a channel for calcium release. When InsP3 is generated in the cell, in response to an external stimulus, it binds to the InsP3 receptor and releases calcium from internal stores. They address InsP3 receptor function in the model organism Drosophila using genetic, molecular, cellular, electrophysiological and behavioural techniques (Srikanth and Hasan, 2004; Banerjee et al, 2006; Kain et al, 2008).

Another colleague in the group, Dr. Mathew has shown that the biological membrane is essentially impermeable to polar and charged solutes. This allows the cell to make separate compartments to carry out different functions. However, it also means that cells need specific proteins to transport these solutes across membranes. They study a variety of transporters in systems ranging from plants to nerve cells in an attempt to understand what these proteins look like and how their structures facilitate the functions they perform. Electrical signaling in nerves is brought about by the movement of ions across the cell membrane. They use a combination of molecular biology and electrophysiology to investigate the mechanism by which the voltage-gated K^+ -channel opens and closes in response to changes in transmembrane potential (Chanda et al, 1999; Varshney et al, 2002; Varshney and Mathew, 2003; Sade et al, 2004).

Dr. Sane, combines the input from physics, engineering, biomechanics, neurobiology, muscle mechanics and behavioural biology to address diverse flight-related phenomena. Specific projects in the lab include the experimental study of fluid dynamics of flapping flight, the role of antennal mechanosensory input in flight control and the integration of multi-sensory inputs in flight behaviour. They are keen to extend the lab studies to the field to study flight energetics, thermoregulation and behavioural ecology of migrating insects. They use diverse techniques such

as a high-speed videography, behavioural measurements, neuroanatomy and neurophysiology to address these questions. They are also interested in the theoretical modeling of various aspects of insect flight at all levels of integration (Lehmann et al, 2005; Sane et al, 2007).

Working at NBRC, Manesar, Dr. Adithya Murthy and his colleagues have revealed aspects of vision neurophysiology, not known earlier. It has been pointed out that before each gaze shift, perceptual processing must identify potential targets for the eye movement, convey this information to higher centers in the brain where decisions to prepare and execute the motor command occur. Their research addresses the challenging question of how information processed by the visual system guides the motor systems to produce overt behavior, spans across the fields of visual perception, attention, and the generation of motor behavior involving the application of cognitive/psychophysical, neuropsychological and physiological techniques in human and non-human primates. (Murthy, 2003; Schall et al, 2003; Camalier et al, 2007).

Dr.Neeraj Jain and his group, also working at NBRC, have been focussing their attention to understand normal and altered organization of somatosensory systems of primates, using more recent techniques and approach. It is pointed out that to generate a coherent sensory percept the somatosensory system of primates processes information from more than 14 different kinds of sensory receptors in the skin, muscles and joints. This information is processed in the brainstem, thalamus, and in more than 7 different areas in the cortex. All these structures are interconnected in a complex feed forward and feed back network, and the information flow is tightly integrated with the motor system. For example, spinal cord injuries in adult monkeys result in somatosensory reorganization of the topographic map in area 3b. The region of the map that normally processes sensory information from the hand now receives sensory inputs from the face. Lack of normal information flow results in changes in the organization and information processing in the brain. The parts of the system that no longer receive sensory information get recruited to process inputs from the remaining intact pathways such as those from the face (Jain et al, 2000b Jain et al, 2001; Jain, 2002; Jain et al, 2003).

Cellular Excitability

The group led by Prof.S.K.Sikdar at IISc, Bangalore has been trying to understand the basic mechanisms underlying the functioning of neurons. They approach this problem using primary cultures of rat hippocampal neurons, cortical neurons and dorsal root ganglion neurons. Recombinant ion channels (Na^+, K^+) expressed in Chinese Hamster Ovary (CHO) cell line are used to understand the functional aspects of ion channels. The modes of approach to address the questions include Patch-Clamp Electrophysiology and Live-Cell imaging. Experiments that involve simultaneous electrophysiological and image acquisitions are carried out.

Combining Electrophysiology and Calcium imaging in understanding the Ca^{2+} dynamics within a solitary neuron their primary interest is in studying the spatio-temporal profiles of calcium dynamics in autaptic hippocampal neuronal cultures. They adopt the microculture approach containing a solitary neuron for the study. Primary cultures of the rat hippocampal neurons are maintained for 1 to 2 weeks in culture after which the experiments are carried out. (Sikdar and Oomura, 1985; Akaike et al, 1987; Sikdar et al, 1989; Tiwari et al, 1996; Tiwari and Sikdar,1999; Majumdar et al, 2004; Senthil Kumar et al, 2005; Harinath and Sikdar, 2005; Majumdar and Sikdar, 2006; Padmashri et al, 2006; Nayak and Sikdar, 2007; Majumdar and Sikdar, 2007; Padmashri and Sikdar,.2007).

Keeping pace with the transition into molecular neuroscience, Ramakrishna's group established a productive collaboration with the Molecular Biophysics Unit of the Indian Institute of Science through the Jawaharlal Nehru Centre for Advanced Scientific Research and studied conformational changes in the beta amyloid peptide (Aâ) as a result of aluminium toxicity. These studies developed a useful *in vitro* model of the pathological conformation and eventual aggregation of Aâ in the brains of patients with Alzheimer's disease (AD) (Ramesh et al, 1999). This approach was also applied to experiments with the microtubule-associated protein tau and suggested a molecular rationale for neurofibrillary pathology in the AD brain. (Madhav et al, 1996) The novel finding that Borate could reverse pathological conformations in Aâ was an exciting discovery arising from these studies (Ramakrishna et al, 1997). In an *in vivo* rat model of aluminium encephalopathy, it was observed that betaine, an inexpensive edible by product from the beet sugar industry and a precursor molecule in acetylcholine biosynthesis, reversed the toxic effect of aluminium. (Ramakrishna et al,1998). These studies were further confirmed by histological studies on dendritic connectivity in the rat hippocampus (Sreekumaran et al, 2003). These experiments spanning *in vivo* and *in vitro* models replicating some of the molecular events in AD pathology provided novel insights into both the pathological cascade as well as approaches to therapeutic strategies for AD.

Even while keeping pace with the march of molecular neuroscience, this group established an international collaborated with Professor David Smith's group in OPTIMA (Oxford Project to Investigate Memory and Ageing) and initiated cross-cultural studies to develop relevant screening tools for dementia in India. A cognitive screening instrument called the 7- minute screen was translated into Malayalam and validated in Kerala (De Jagar et al, 2008).

Invertebrate Neurophysiology

The late fifties saw the establishment of a new university in the south at Tirupati, where a department of Zoology with post graduate and research studies came into being. This was headed by Prof Pampapathi Rao who returned to India after a post-doctoral fellowship with Theodore Holmes "Ted" Bullock at the University

of California. By then Ted Bullock and Adrian Horridge were well known for their seminal contributions to our understanding of the structure and function of nervous systems in Invertebrates. Carrying forward that legacy in India, Rao initiated similar studies in arachnids, evolutionarily an ancient group of organisms, of the Phylum Arthropoda. Using the scorpion as a model, his group discovered blood-borne factors modulating circadian rhythms of locomotor activity synchronised with the electrical activity in the ventral nerve cord (Rao and Reddy, 1967). A major discovery during this period was the description of proprioceptors in the dorsal leg nerve of the scorpion. This seminal work was reported in Nature in the early sixties (Rao and Murthy, 1966).

Another major breakthrough during this period was the discovery of neurosecretory factor(s) regulating temperature acclimatisation in poikilotherms which was reported by Pampapathi Rao in the journal, Science (Rao, 1962).

While using an invertebrate as a model for obvious reasons of relative simplicity of the nervous system in terms of the small number of neurons and the limited patterns of the behavioural repertoire generated by them, the ultimate goal of understanding complex behaviour in man was never lost sight of. A further high point came in early seventies, when, through the application of cybernetic principles, it was shown that circadian rhythms are "state-determined " and are in synchrony with non-specific sensory input, differentially during active and quiescent phases. It must be mentioned that the application of the rigour of mathematics/ cybernetics to these experiments was very much ahead of its time. This work was reported in the proceedings of the Indian Academy of Sciences B (Ramakrishna and Rao, 1971).

Yet another fundamental contribution came from this group during the late seventies, when, using glass micro-pipette electrodes, depth recordings of the electroretinogram (ERG) in the scorpion were performed to establish the dipole nature of the light receptor (Ramakrishna, 1977; Ramakrishna, 1983). A closely related and significant contribution was the discovery of a metasomatic neural photoreceptor in the scorpion by Geethabali and Rao from the electrophysiology laboratory of Bangalore University (Geethabali and Rao, 1973). In later decades several aspects of drosophila mutants, gens involved in regulation of neurons, electrical responses of taste receptors of x-linked gene mutants of drosophila, and axonal counts of all prominent nerves of mouth parts have been described (Singh and Nayak, 1985). The studies have been continued with focus on genetic, cellular, molecular and behavioural levels of using Drosophila as a model (Kain et al, 2008).

Oscillometry and Biopotentials

An entirely new approach and method has been pioneered by Mendanha and his colleagues at Pune in attempting to record high and ultrahigh frequency domain of biopotentials, a field virtually unknown till recently. Use of oscillometry – a method developed by Mendanha, is easy in operation, and is based on simple

electronic and physics principles. Raw full spectrum signals are fed into the stereo line-in input of a sound card with a sample rate of 96 KHz. The analogue signal is digitally recorded. The power of the signal in decibels can be measured. The spectral view display shows the moment to moment changes in frequency content of the signal. The Fast Fourier Transform (FFT) resolves frequencies from 0 to 45 KHz. The method permits unlimited recording time and computerized analysis. Mendanha has been using the method to study energy flows and with suitable protocols have been used to test high frequency biopotential changes observed in yogis during yogic practices (Mendanha, 2001, Mendanha, 2008). It has been proposed that using more sophisticated multichannel electrical recording devices with greater stability in the extremely high frequency range is likely to broaden the scope of the studies (WHO, 1999).

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