REVIEW ARTICLE

AUTONOMOUS NEURAL INFLAMMATORY REFLEX AND CONTROL OF INNATE IMMUNITY: TOWARD INNOVATIVE TREATMENT OF UNCONTROLLED INFLAMMATION

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Background
Inflammation is common pathology associated with infections and other diseases process that lead to non specific sickness behaviours. Identification of autonomous neural inflammatory reflex that is regulated through autonomic nervous system and their receptors give a way forward on how this can be use as therapeutic measures in the treatment of uncontrolled inflammatory disorders.
We review the available articles within our reach on neuroimmunology and its role in regulating the inflammatory response in disease process with emphasis on possible therapeutic modalities.
The nervous system reflexively regulates the inflammatory response to infectious agents and in other disease process, cytokine networks and neural autonomic pathway through vagus nerve monitor inflammatory status and coordinate appropriate host defences. Immunomodulatory stimulation of vagus nerve, acetylcholinesterase inhibitory mechanism and 7 nAChR receptor expressed on immune cells plays a role on autonomous neural inflammatory reflex. This can be therapeutically applied in the treatment of inflammatory uncontrolled disorders.
Aim of the study: *To review developing concept of neurological role in the regulation of innate immunological reaction to infections in relation to autonomic nervous system and highlights the therapeutic application of the concept. To understand the concept of immunomodulation in management of certain inflammatory disorders.*
keywords: *Autonomous, Inflammatory reflex, vagus nerve, immunomodulation, therapeutic modalities*

Introduction

Reflex control of immunity is a new concept following advances in neuroimmunology. All the five sense organs in humans and animals as well as response to infective agent are controlled by neural stimulation[1]. Infectious agents are associated with pathogen-associated molecular patterns (PAMPs) which are linked by pattern recognition receptors (PRRs). The PRRs are expressed on the surface e.g. Toll-like receptor (TLR4) and inside the cytoplasm e.g. nucleotide-binding oligomerization domain (Nod) like receptors of cells of the innate immune system, primarily macrophages and dendritic cells. The cytokines and bacterial toxins deliver information to the brain using both humoral and neuronal routes of communication through binding to PRRs and activation of vagus nerve. All brain cells are capable of secreting cytokines following neural and humeral stimulation by an infective agents. Inhibition of pro-inflammatory cytokines that are induced following bacterial infection blocks the appearance of sickness behaviours.

Innervation Of Immunity Of The Immune System

Nerve fibres are involved in local monitoring and modulation of host defence coordinated by Central Nervous System (CNS). Spleen has an organisation of nerves in the white pulp, separated from the red pulp by the marginal zone. Splenic macrophages production of tumour necrosis factor (TNF) in the red pulp and marginal zone is regulated by vagus nerve signals that are transmitted through coeliac ganglion (fig. 1). Cells of the immune system pass through the vascular matrix of spleen in close proximity to nerve endings arising from the splenic nerve. Electron microscopy and immunohistochemistry, revealed nerve endings in the vicinity of T cells, B cells and macrophages which originate from brain stem, sympathetic chain, and peripheral ganglion cells. Nerve Synapses were found lying within about nanometers of immune cells that express receptors for acetylcholine, catecholamines, neuropeptides, and other neurotransmitters[2,3].

Inflammatory Reflex

Inflammatory reflex (Fig. 1) is a typical neural circuit, activation is by exogenous and endogenous molecular products of inflammation through afferent action potentials traveling in the vagus nerve to the nucleus tractus solatarius and relays the neuronal signals to other brain nuclei located in the hypothalamus and brainstem[2,3]. The efferent signals travel from the nucleus ambiguus and dorsal motor nucleus back to the vagus nerve and terminates in the coeliac ganglia[4]. Stimulation of the vagus nerve results in the activation of adrenergic splenic neurons of the coeliac ganglion, which travel into the spleen and terminate in synapses in the white pulp adjacent to T cells[5]. Splenic neurons releases norepinephrine which binds to adrenergic receptor expressed on a subset of T cells that expresses choline acetyltransferase. Acetyltransferase act as the rate-limiting enzyme in acetylcholine biosynthesis. Acetylcholine that acts as the terminal neurotransmitter is provided by T cells in the inflammatory reflex circuit. Nerve endings of the spleen synapses in the splenic white pulp and expresses adrenergic receptors which respond to

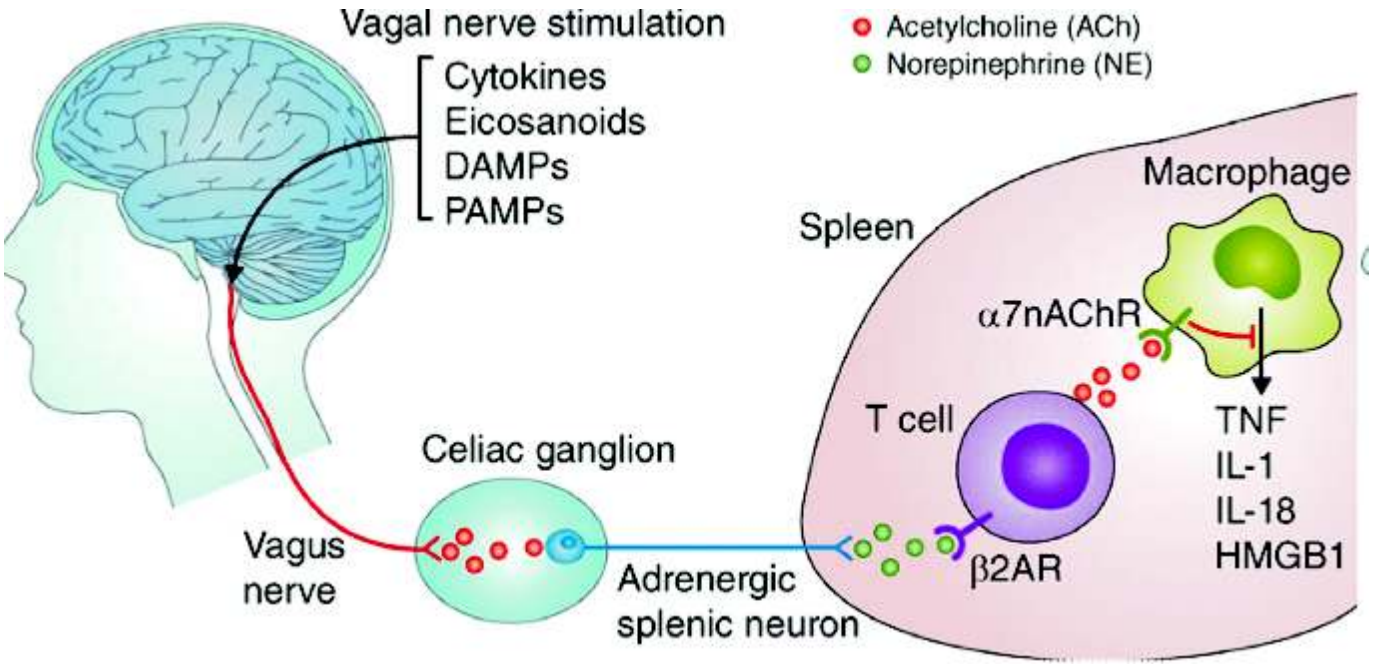

Fig 1: Vagus nerve and regulation of cytokines production. Source: Neural reflexes in inflammation and immunity. Ulf Andersson and Kevin J. Tracey; JEM 2012.

T cells that produces acetylcholine are also found in lymph nodes and Peyer's patches[5]. Acetylcholine binds to 7 nAChR receptor that is expressed on macrophages in the red pulp and marginal zone[5] and the generated signals that down-regulate cytokine synthesis by suppressing nuclear translocation of NF-B. This has inhibitory activity that dampens the activity of the innate immune response to pathogen associated molecular products[5]. This inhibitory activity can be enhanced by methods that increase the generation of adrenergic signals in the splenic nerve through electrical stimulation of the vagus nerve or splenic nerve and directly by pharmacologically activating the adrenergic splenic neurons using cholinergic agonists[6]. The efferent vagal signal significantly enhances spleen norepinephrine and acetylcholine release, and suppresses cytokine production[6]. This promote utilization pharmacological agonists that target 7 nAChR to deactivate macrophages as well as pharmacological agonists that target muscarinic acetylcholine (M1) receptors in the brain to up-regulate efferent vagus nerve activity. Pharmacological M1 agonist that activate vagus nerve suppresses inflammation in vivo[6,7]. It was found out that patients with autoimmune disease and non-resolving inflammation have profound impaired vagus nerve signaling, which accelerates the progression of inflammation and prevents resolution[8]. Migration of leucocytes through 7 nAChR to inflammatory area is regulated by vagus nerve activity in the spleen and treatment with 7

nAChR agonists or electrical vagus nerve stimulation significantly reduces the inflammatory symptoms[9]. There was reported increased mortality rate in severe sepsis of up to 60% in patients with decreased vagus nerve activity as compared with 0% mortality in subjects with higher vagus nerve activity[10]. Vagotomy following accidental injury is associated with significant increased in mortality and septicemia as compared with a cohort of injured control patients[11]. There was also reported reduced vagus nerve activity in rheumatoid arthritis patients compared with controls[12]. Vagus nerve deficiencies can be reversed by vagus nerve stimulation through administration of 7 nAChR agonists, or by physiological methods that enhance vagus nerve activity, including aerobic exercise, acupuncture, meditation, music therapy, and biofeedback training.

Dietary anti inflammatory reflex[13] (Fig. 2) operate in similar passion through cholecystokinin (CCK) receptors in which consumption of dietary fat is associated with inflammatory reflex response. It was reported that dietary consumption of fish oil enhances the basal vagus nerve activity acting as anti-inflammatory mechanism and enhance resolution of inflammation[14]. There is also an immunosuppressive stroke reflex**,** this is due to the fact that stroke survivors were found to have an associated increased susceptibility to spontaneous bacterial sepsis and pneumonia due to adrenergic mediated extensive lymphocyte apoptosis, and shift in cytokine production from a T helper cell type 1 to T helper cell type 2 series[15,16]. The associated risk of infection was significantly reduced after pharmacologically blockading alpha-adrenergic receptors with propranolol[15,16].

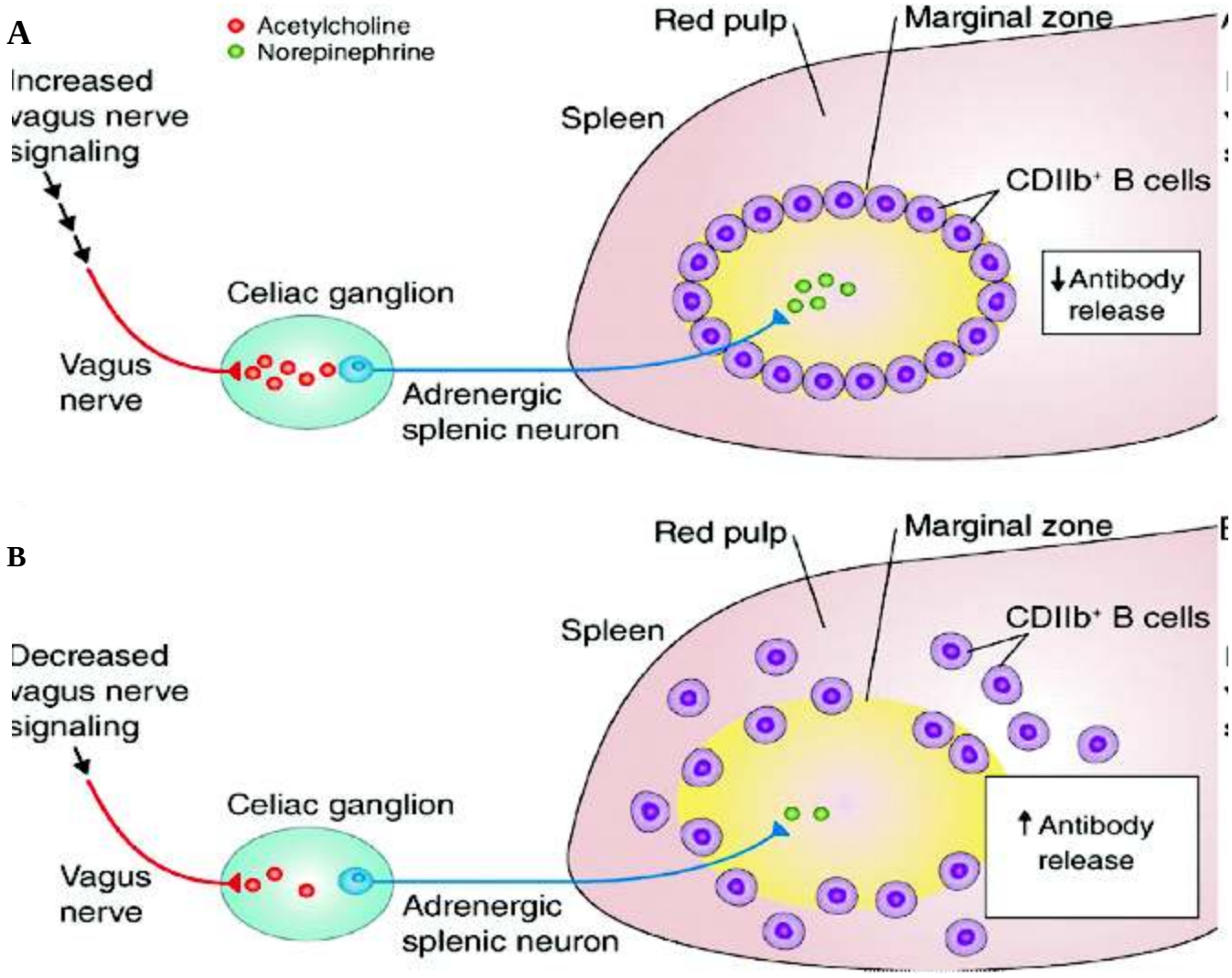

Figure 2: Dietary antiinflammatory reflex.
Neural reflexes in inflammation and immunity. Source: Ulf Andersson and Kevin J. Tracey; JEM 2012

Neural Control Of Antibody

Production

Mechanisms B cell trafficking during maturation into antibody-secreting cells is also regulate by autonomic nervous system. Phosphorylcholine in the spleen initiate the antibody response and result in the accumulation of antibody-secreting B cells in the red pulp which releases antibodies into the splenic venous circulation. Electrical stimulation of vagus nerve or administration of nicotine impaired migration and transformation of B cells into plasma cells, they accumulate in the marginal zone which reduces antibody secretion[17]. This neural regulation of B cells transformation to plasma cells prevent excessive production of antibody and influence the nature of the adaptive immune response.

Therapeutic Application Of Neural Reflex Control Of Immunity

Specific steps in immunity are regulated by neural circuits that signal through defined molecular mechanisms reflexively. Cholinergic anti-inflammatory pathway regulate immune responses and the progression of inflammatory diseases. This is important as a therapeutics modalities that target neural networks for the treatment of inflammatory disorders. Advances in the fields of biomedical and electrical engineering enabled development of new neuro-stimulators (Table 1) that can be implanted selectively in the brain or along the course of the vagus nerve or other nerves to modulate the activity of neural circuits. Controlled electrical modulation of vagus nerve activity inhibits cytokine release and confers considerable protection from tissue injury in arthritis, sepsis, inflammatory bowel disease, ischemia-reperfusion injury and ileus. Neurotransmitters and their associated receptors were also used in the development of new pharmaceuticals agents to control immunity.

Neuromodulation

Neuromodulation is the physiological process by which a given neuron uses one or more neurotransmitters to regulate diverse populations of neurons, the neurotransmitter cannot be reabsorbed by the pre-synaptic neuron or broken down into a metabolite. The neuromodulators spend a significant amount of time in the cerebrospinal fluid (CSF), modulating the activity of several other neurons in the brain (Table 1). The receptors for neuromodulators are typically G-protein coupled receptors while in classical chemical neurotransmission are ligand-gated ion channels. G-protein linked receptors of neuromodulators involves voltage-gated ion channels, and is relatively slow while neurotransmission that involves exclusively ligand-gated ion channels is much faster.

IMMUNOMODULATORY DRUGS

Thalidomide

Thalidomide and its analogues lenalidomide and Actimid are immunomodulatory drugs with anti-angiogenic properties, they modulate TNF alpha and IL-12 secretion, co-stimulate T cells, increase NK cell toxicity and have direct anti-tumour effects[18]. Lenalidomide and Actimid are more powerful in inhibiting TNF alpha and have less side effects than Thalidomide.

Avipaxin

Avipaxin (HupA) is a potent, highly specific, and reversible acetylcholinesterase (AChE) isolated from the Chinese herb *Huperzia serrata*. The AChE is primarily localized in the central nervous system supporting HupA's selective increase of acetylcholine in the brain[19]. Avipaxin's, immunomodulatory effects are dependent on CNS muscarinic activity associated with alpha 7 nicotinic acetylcholine receptors (α7nAChRs). When activated by binding of acetylcholine, lead to decreased synthesis and release of cytokines. This immunomodulatory effects of HupA are also dependent on the vagus nerve and the splenic nerve. Inhibition of acetylcholinesterase by Avipaxin lead to sustain acetylcholine causing sustain activation of vagus nerve and dampen uncontrolled inflammation. The vagus nerve modulates immune system activation indirectly by stimulating the splenic nerve, which innervates the spleen[20]. Upon stimulation of the descending vagus nerve from the brain, acetylcholine is released at the coeliac-superior mesenteric plexus ganglion causing activation of the splenic nerve. The splenic nerve innervates the spleen and releases catecholamines, leading to attenuation of cytokines by binding to β-adrenergic receptors. In addition, it has been proposed that catecholamine release from the splenic nerve can enhance acetylcholine levels in the spleen, which can act on α7nAChRs to attenuate cytokine production.

Table 1: Various experimental neuromodulators agents and their therapeutic applications

EXPERIMENT	ACTIVATION	RESULT
Endotoxin induced shock	Direct electrical stimulation of vagus nerve	Inhibition of serum TNF secretion and attenuation of shock
Sepsis induced by lethal peritonitis	Transcutaneous stimulation of vagus nerve	Inhibition of serum HMGB-1 secretion and improved survival
Pancreatitis	Administration of α7nAChR against GTS	Inhibition of serum HMGB-1 secretion and improved survival
	Vagotomy and administration of α7nAChR antagonist	
Inhibition of serum HMGB-1 secretion and improved survival	Direct electrical stimulation of vagus nerve	Inhibition of neutrophils recruitment to the wound
colitis	Vagotomy and administration of α7nAChR antagonist	Increase colitis severity
	administration of α7nAChR agonist	Decrease colitis severity

Discussion

Sympathetic role in immunological disease processes was found to be associated with chronic stress, depression and aging[21]. Inappropriate stimulation of sympathetic catecholamine levels due to the brainstem aseptic trauma like head injury lead to catecholamine-induced systemic immunosuppressive cytokine profiles which lowered TNF-alpha production and down regulate monocyte antigen presentation with subsequent increased in the frequency and severity of post-operative infectious complications. This catecholamine-stimulated release of monocyte IL-10 appears to be rapid and direct, without involvement of immunological costimulation.

The body manages to respond to infectious agents through sentinel cells such as kupffer cells, langerhans cells and alveolar macrophages located throughout the body with common set of symptoms despite a lack of similarities between these types of pathogens. This response of the innate immune system results in behavioral consequences associated with the neuronal activity and the behaviours returns to normal after resolution of the inflammation. In a severe form of neuroinflammation, prolong cell death within the CNS can occur leading to irreversible loss of function and unresolved inflammation.

Recognition of the infectious agent is associated with activation of neural and humoral routes that provide input to the brain and elicit behavioural response that are dependent on the site of infection. Vagotomy drastically reduces the sickness response to intaperitoneal lipopolysaccharides[22] with associated increase IL-1 levels within the brain[23,24] but does not block the pyrogenic action of LPS when LPS is administered[25]. Additional humeral activity is required to illicit this sickness behaviour because vagotomy does not block the induction of this behaviour after intravenous or subcutaneous LPS[26]. The behavioural responses that occur in response to intravenous and subcutaneous PAMPs or cytokines are transcribed by the brain in response to humoral signals and some of the behaviors that occur in response to intraperitoneal challenges have a humoral component even after vagotomy[23,25]. All

behavioural responses to infection have a cytokine basis and LPS induced CNS inflammatory response correspond to *c-fos* activation by the vagal nerve afferent projections[27]. Intraperitoneal or meningeal infections associated behavioural changes are partially mediated by neural afferents through the vagal and trigeminal nerves respectively while other peripheral sites of infection have a stronger dependence on the humoral pathway.

IL-1 β from periphery delivered to CNS by humoral pathway or expressed within the brain or exogenously added is dependant on neural pathway to induce sickness behaviour[28,29]. This IL-1β receptor pathway is predominant in monocytes, including brain microglia and the activation lead to elevated cytokine expression, further monocyte/microglia activation and astrocytes activation within the CNS[30].

TNF alpha expression within the brain is also dependent on neural input that act through TNF-R1 that are primarily localised to neutrons in the brain to induce sickness[31].

Recommendations

More therapeutic evaluation trial and physiological studies of immunomodulators need to be done to warrant the utilization of such agent on patients as a therapeutic measure to improve the outcome chronic inflammatory disorders and sepsis.

Conclusion

The autonomic nervous system reflexively regulates the inflammatory response to infectious agents which provide an insight to the treatment of chronic inflammatory process through selective and reversible neural reflex pathway of control of inflammation. Cytokine networks as regulated by neural autonomic pathway monitor inflammatory status and coordinate timely appropriate host defences. This information on signal transduction, role of other neurotransmitter receptors and advances in the field of antibody-mediated effects on brain function challenges immunologists to try various therapeutic modalities in the treatment of refractory inflammatory diseases and sepsis.

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