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Review article

Comprehensive biology of antipyretic pathways

Prajitha N, Athira SS, Mohanan PV*

Toxicology Division, Biomedical Technology Wing, Sree Chitra Tirunal Institute for Medical Sciences and Technology, Poojapura, Trivandrum 695 012, Kerala, India

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ABSTRACT

Pyrogens, the fever inducing substances accidently enter into a human body through contamination from medical or pharmaceutical products may create mild to severe complications including septicaemia and shocking syndromes. To avoid such drastic situations all the pharmaceuticals and medical devices are analysed for presence of pyrogens prior to their release into market. The entry of exogenous pyrogens like bacterial endotoxins induces the release of endogenous pyrogens or inflammatory cytokines that activate immune system to defend against these pathogens. Generation of heat is considered as one of the important defence mechanism of body achieved through receptor mediated interaction of endogenous pyrogens at the thermoregulatory centre of hypothalamus. However, uncontrolled fever and febrile reaction may cause lethal effects to the subject itself. So a well sophistically functioning antipyretic mechanism is necessary to achieve thermoregulation. The co-ordinated interaction of antipyretic cytokines and other mediators are active in human immune system which play a crucial role in maintaining thermal homeostasis. The multiple interacting antipyretic signals and their mechanism are the major subjects of this review.

1. Introduction

The word 'antipyretic' literarily means the agents that reduce fever but have no effect on normal or physiologically increased body temperature. Normally, the thermal regulatory network of the body maintains a temperature of 36.2–37.5 °C [1] by various endogenous mechanisms; to prevent brain damage and helps to maintain thermal homeostasis via regulated functioning of Central Nervous System (CNS) to avoid a life threatening conditions. Both peripherally and centrally active mechanisms are involved in endogenous antipyresis. Peripheral endogenous antipyretic action is mediated by antipyretic cytokines, glucocorticoids; Spleen derived anti-hyper pyretic lipid factors and several other hormones. Centrally active antipyretics include eicosanoids, neuropeptides, nitric oxide and other gaseous neurotransmitters. During the course of fever, endogenous antipyretic mechanism remains elaborated within the body to limit both strength and duration of fever mainly through inhibition of enzymes necessary to synthesize inflammatory mediators. Although fever and febrile reactions are the initial defence mechanism of our body, persistent fever for longer duration will be detrimental. Hence, several pharmaceutical drugs are introduced to restore the well-being from sickness behaviour associated with pyrogens. Most of these antipyretics are group of drugs called non-steroidal anti-inflammatory drugs (NSAIDs) [2].

2. Endogenous antipyretic cytokines

Pyrogens are agents which induce fever in humans and are either external or internal in nature. The entry of exogenous pyrogens like bacterial endotoxins and associated release of endogenous pyrogens including Interleukin-1β (IL-1β), Interleukin-6 (IL-6) and Tumour necrosis factor-α (TNF-α) induce fever and other inflammatory response within the body and eventually result in loss of homeostasis of the immune system (Fig. 1). To counter balance the effect of released pro-inflammatory cytokines, there exist several antipyretic mechanisms. It involves the production of anti-inflammatory cytokines, internalization of endotoxins-receptor complexes, down regulation of cytokine - receptor complexes [3], release of soluble receptors for Interleukin-1 (IL-1), Tumor necrosis factors (TNF) and release of antagonists ofIL-1 and its receptor (IL-1ra) [4]. Major anti-inflammatory cytokines in action are: IL-4, IL-10, IL-11, IL-13, IL-18 and TNF-β [5].

2.1. Interleukin 10 (IL-10)

Among the group of anti-inflammatory cytokines, IL-10 is considered as the most active antipyretic cytokine with inhibitory action on production of TNF, IL-1, IL-6, IL-8 and up regulating action on expression of IL-1ra [6]. Cartmell et al., in 2003 observed that injection of Lipopolysaccharides (LPS) isolated from pathogenic bacteria like *E.coli*

* Corresponding author.
E-mail address: mohanpv@sctimst.ac.in (M. PV).

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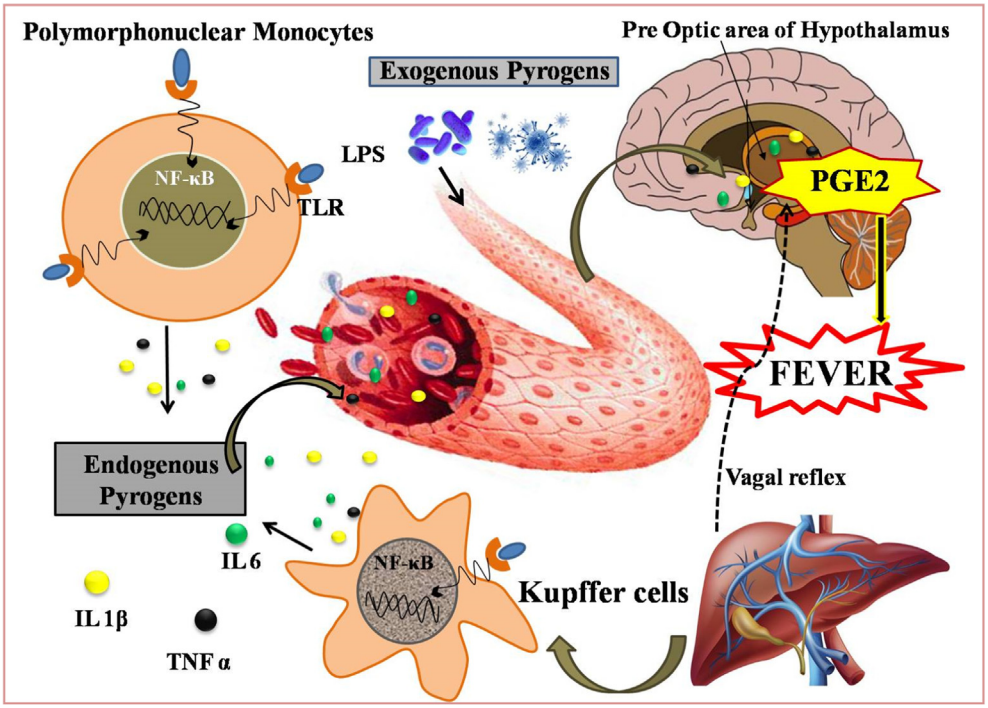

Fig. 1. Induction of fever by Pyrogens.

and *S.aureus* toxins into subcutaneous air pouch of rat suffering from intense fever and inflammatory reactions. Administration of LPS evoked fever that began approximately 2 h later injection reached maximum at 3 h and persisted up to 8 h. Similarly, administrations of S. aureus toxin also evoked fever approximately 2 h later injections; with an initial peak of 38.9 ± 0.2 °C at 6 h. Administration of rat recombinant IL-10 along with these bacterial toxins significantly reduced the magnitude of fever and febrile reactions. However, recombinant IL-10 has no profound inhibitory effect on concentration of pro-inflammatory cytokines like IL-1β and TNF-α within the animal. Treatment with sheep anti-IL-10 reduced the level of IL-10 evoked by LPS and *S. aureus* toxin in air pouch. Anti IL-10 administration caused increased production of local (pouch) IL-6, but showed no effect on local and circulating concentrations of TNF-α, IL-1β and IL-1ra, and circulating IL-6 concentration, evoked by LPS 5 h after injection. However, it potentiated the increases in TNF-α, IL-1β, and IL-6 at the site of *S. Aureus* injection; and increased circulating concentrations of IL-6 and IL-1ra after 5 h of *S. aureus* injection [7]. Leon et al., (1999) agreed with similar results from their experiment using healthy Swiss Webster (SW) and IL-10 knockout mice. Fever and febrile reactions after intraperitoneal administration of *E.coli* LPS was found to be reduced by the presence of IL-10 in SW mice, whereas only 30% of IL-10 knockout mice was survived after injection of high dose of LPS (2.5 mg/kg). Unfortunately, IL-10 failed to reduce fever induced by local inflammatory agent like turpentine [8]. In the same way, Harden et al., (1999) selected LPS stimulated Organum Vasculosum Laminae Terminalis (OVLT) and area postrema (AP) primary cells isolated from rat pup brain tissue to analyze the modulatory action of IL-10 on the synthesis of pro-inflammatory cytokines [9]. By incorporating rat IL-10 antibodies (30 μg/ml) into the primary cell culture, it was observed that inhibition of IL-10 enhanced the production of TNF-α and IL-6 from OVLT and AP cells following LPS stimulation.

Stimulation of IL-1ra production, an alternate function of IL-10 was analyzed by measuring the concentration of IL-1ra and their mRNA expression within cultured polymorphoneutrophil cells (PMN) using Enzyme Linked Immunosorbent assay (ELISA) and northern blotting techniques. From the experiment, Crepaldi et al., (2002) reported that IL-10 have an important role in the up regulation of IL-1ra synthesis from PMN stimulated prior to IL-4 [10]. Conceptually, when macrophages are infected by LPS, they require metabolic energy renewal via active uptake and cleavage of glucose. IL-10 has negative effect on glucose uptake mediated by GLUT1 transporters and also reduces the expression of enzymes of glycolytic pathway. IL-10 also shows inhibitory effect on caspase-1 dependent inflammasome activation so that the production of IL-1β become defective in LPS treated macrophages. Thus, from this experiment Ip et al.,(2017) well concluded that IL-10 exhibits anti-inflammatory activity chiefly by inhibiting mammalian target of rapamycin (mTOR) signalling pathway necessary for the metabolic rearrangement of cells during an infection or inflammatory circumstances[11].

3. Antipyretic hormones

3.1. Glucocorticoids

The hormone glucocorticoids, released from adrenal cortex have anti-inflammatory and immunosuppressive properties which can be attributed to the transcription inhibition of inflammatory cytokines and chemokines including IL-1α, IL-1β, IL-2, IL-3, IL-4, IL-5, IL-6, IL-11, IL-12, IL-13, IL-16, IL-17, interferon-γ (IFN-γ), TNF-α,CC-chemokine ligand 2 (CCL2), CCL3, CCL5, CCL11, CCL24, CCL26 and granulocyte–-macrophage colony-stimulating factor (GM-CSF)[12]. Presence of stress induced fever in adult adrenalectamized rats after neonatal LPS exposure was found to be an indication of the role of antipyretic action of corticosterone. Corticosterone influenced the release of cyclooxygenase (COX) enzymes especially COX2 and subsequent production of prostaglandin E2 (PGE2) frompre-optic areaof hypothalamus (POA) in adrenalectamized rats [13]. Hypothalamic-pituitary-adrenal (HPA) axis plays an important role in immunoregulation via co-ordinating the signals from CNS and endocrine system. Elevation of both free and bound form of corticosteroids followed by intraperitoneal injection of LPS to rats indicates their presence in controlling pyrexia associated with endotoxin infection [14].

Similarly, there occurred Nuclear factor kappa B(NFκB) induced gene expression and release of IL-6 in rats after LPS (50 μg/kg) challenge and the response was attenuated by the presence of

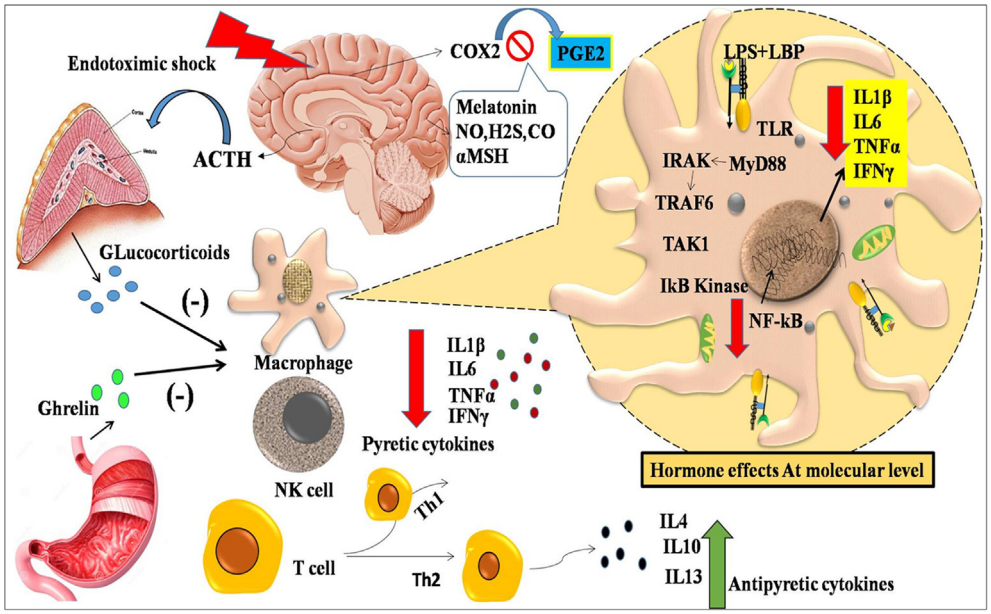

Fig. 2. Antipyretic effect of Hormones.

corticosterone. However, administration of glucocorticoid receptor antagonist; RU-486 increased the magnitude of fever and febrile reactions. Notably maternal care at infant stage exhibited a positive influence on reducing febrile reactions in adult stage when they are later challenged with LPS [15]. Thus, these studies provide strong evidence that endogenous glucocorticoids have physiological antipyretic activity. It was further confirmed that the classic stress and LPS induced response of glucocorticoids function as a counter regulatory modulator of febrile responses.

3.2. Ghrelin

Ghrelin, an orexigenic 28 amino-acid peptide, secreted by gastric oxyntic mucosa has role in controlling appetite and energy homeostasis. Also ghrelin acts as an endogenous ligand for the growth hormone (GH) secretagogue receptors (GHS-R) and promotes GH dependent immune modulation. A recent study implicated the putative role of ghrelin in LPS induced fever. Exogenous administration of ghrelin (0.1 mg/kg) along with intra peritoneal (i.p)injection of LPS (50 μg/kg) significantly reduced fever and febrile reactions in rats. There observed associated rise in plasma corticosterone after injection. From this experiment Soriano et al., (2011) confirmed that the production of PGE2 after LPS induced febrigenicthermo-effector mechanism was altered in presence of ghrelin [16]. Likewise, Wang et al., (2006) found that there is a correlation between i.p injection of LPS (100 μg/kg) and circulating level of ghrelin in rats. Both LPS and IL-1β seemed to suppress the production of ghrelin wherein IL-1ra over whelmed the effect [17]. Ghrelinalso exhibits direct effects on defence function of macrophages through interaction with GHS-R *in vitro*. Pre-treatment of RAW264.7 macrophages and SN4741 neurons with exogenous ghrelin prior to LPS stimulation resulted in dose dependent inhibition of translocation of p65 subunit of NFκB necessary for the active transcription of progeny cytokines including IL1-β and TNF-α. In addition, ghrelin also augmented the production of antipyretic cytokine IL-10 in a dose and time depended manner through its target growth hormone secretagogue receptors present in macrophages [18]. It was also reported that, introduction of LPS challenge to SN4741 dopaminergic neurones in mouse enhanced the secretion of IL-6 and was significantly attenuated by ghrelin within 24 h. However in neuronal cells, nuclear translocation of NFκB has no adverse effect through ghrelin interaction like seen in macrophages [19].

3.3. Melatonin

Melatonin; hormone secreted by pineal gland plays important role in regulation of circadian rhythm and is known to possess several other physiological functions like free radical scavenging, anti-apoptotic, visual, reproductive, cerebrovascular, neuroendocrine andneuro-immunological actions. The anti-inflammatory action of melatonin was analysed by analyzing inflammatory stress induction through LPS exposure in neuronal stem cells (NSCs). LPS treatment affected the viability of NSC and induced inflammatory damages. LPS enhanced apoptotic cell death and decreased number and size of neurosphere around the cells. There observed slight decrease in mRNA levels of SOX2, FGFR-2 (markers of NSC survival and proliferation) and expression of PI3K, Akt and Nrf2 (Proteins of cytoprotection pathway) in LPS treated cells compared to untreated control. Presence of Melatonin improved the survival of NSC cells through reducing membrane destruction and NO mediated oxidative stress response. Melatonin treatment helped to maintain the neurosphere size during LPS mediated inflammatory condition and also enhanced the expression of SOX2 and FGF2 mRNAs. There occurred a little increase in expression of PI3K, phosphorylated-Akt and Nrf2 proteins after melatonin treatment during LPS inflammation. Hence, it was noted that melatonin has beneficiary effect to survival of neuronal cell during inflammatory circumstances [20].

Apart from reducing the expression of IL-1β, TNF-α, IL-6, GM-CSF, CCL2, CCL5, serum amyloid A, haptoglobin, C-reactive protein, ceruloplasmin, α-1 antitrypsin in LPS treatedbovine mammary epithelial cells [21], melatonin inhibited acetylcholine esterase activity and MDA level in different brain regions (striatum, cerebral cortex, hippocampus and hypothalamus) of rat challenged with LPS. Melatonin also enhanced the production of antioxidant like reduced glutathione *in vivo* [22]. Role of melatonin in liver injury associated with D-Galactosamine/LPS administration in mice was assessed by Wang et al., (2007) and reported that melatonin gave protective effect in hepatic system. Along with activation of anti-oxidant enzyme systems in liver, melatonin significantly reduced TNF-α and activation of apoptotic enzyme caspase 3 in hepatic cells [23]. Summarized antipyretic effects of hormones are depicted in Fig. 2.

4. Interaction of antipyretic mediators in hypothalamus

4.1. Neuropeptides: -Melanocortins

Melanocortin is the posttranslational product of pro-opiomelanocortin (POMC) prohormone. POMC is the precursor of α, β, and γ-melanocyte-stimulating hormone (α,β,γ-MSH) and adrenocorticotropic hormone(ACTH). They interact with immune cells via G protein-coupled melanocortin receptors (MCR). Melanocortins are involved in diverse number of physiological functions including pigmentation, steroidogenesis, energy homeostasis, exocrine secretion, sexual function, analgesia, inflammation, immune-modulation, temperature control, cardiovascular regulation and neuromuscular regeneration [24]. Tatro et al., (2000) reviewed in detailabout thestudies related to antipyretic effect of α-MSH in animal model which deals with the consistent thermoregulatory effects of α-MSH especially on CNS. Also the review highlighted the regulation of LPS induced fever by endogenous melanocortin chiefly through receptor mediated signalling mechanism [25].

Clark et al., (1985) analyzed the antipyretic activity of α-MSH compared to the drug acetaminophen. α-MSH was introduced via intra cerebro-ventricular (i.c.v) injection into rabbits after inducing fever by means of administration of sodium arachidonate (i.c.v.), PGE2 (i.c.v.) or leucocytic pyrogen (i.v.). It was observed that α-MSH (200 ng) exhibited significant antipyretic effect against leucocytic Pyrogens but it neither antagonized PGE2 nor altered arachidonate mediated hyperthermia in rabbits [26]. Likewise, endogenous antipyretic effect of α-MSH was experimented in rats suffering from LPS induced fever. It was found that endogenous melanocortins acts on MCR within the brain to attenuate fever and febrile reactions. Simultaneous *in vitro* experiment with rat melanoma cells showed that melanocortins have specific interaction with their receptors (MCR3 and MCR4) on the surface of melanoma cells which lead to the accumulation of cyclic AMP within the cell. However, the antagonist SHU-9119 inhibited the action of α-MSH both *in vivo* and *in vitro*. In particular, central administration of SHU-9119 to febrile rats exacerbated fever, whereas intravenous administration of the same dose of SHU-9119 had no effect on LPS induced fever. These findings indicate the centrally active antipyretic effect of α-MSH is independent of any modulation in the activity of the pituitary-adrenal axis [27].

4.2. Hypothalamic hormones

Vasopressin is an endogenous neuropeptide or plasma neurohormone synthesized from the par ventricular nuclei within the hypothalamus. Some recent studies have suggested that in addition to vasoconstriction and antidiuresis [28], Arginine vasopressin (AVP) have immuno-modulatory effect in sepsis. In order to elucidate antipyretic action of vasopressin, Peng et al., (2013) and Park et al., (2015) selected RAW264.7 cell line and investigated its role against LPS induced response within these macrophage cells. It was observed that AVP negatively regulated the production of chemokines; TNF-α, IL-1β, IL-6, NO/iNOS mRNA, PGE2/COX-2 mRNA, IKK activity, IκBα degradation and LPS-induced NFκB transcription within the macrophages [29,30]. Similar to LPS immune challenge, i.c.v. administration of IL-6 in rats seemed to have significant role in AVP neuron activation in the supra optic nucleus of hypothalamus [31]. A dose dependent production of AVP after LPS stimulation from supra chiasmatic nucleus slice cultures was reported earlier [32]. Later it was found that LPS acts via interaction with angiotensin II type1 receptors located within the blood brain barrier. Also it was noted that signalling from central angiotensin II is essential for regulation of blood pressure during endotoxemia [33].

Similarly, down regulation of AVP-V3 and corticotropin-releasing factor receptor1mRNA within the anterior pituitary was observed in Holstein steers, during stress response to LPS challenge. In some of these bovine species, AVP and corticotrophin releasing factors showed ACTH releasing activity so that it could indirectly affect the pituitary–adrenal response to stress [34].

5. Gaseous neurotransmitters

5.1. Nitric oxide

Endogenously, the catabolism of amino acid L-arginine by the enzyme NO synthase results in the formation of L-citrulline and Nitric oxide (NO). Three [endothelial (e), neural (n), and inducible (i)] isoforms of nitric oxide synthase (NOS) are known to be expressed in various tissues. NO acts as gaseous neurotransmitter and its interaction increases cyclic GMP levels in tissues, including vascular smooth muscle and CNS. Over the years, NO was demonstrated in many of the pathophysiological functions like thermoregulation and fever. In humans, NO has role in pyretic febrile response by acting at OVLT and POA; both of which constitute the important sites through which circulating pyrogens signals the brain [35]. However in rabbits and guinea pigs, it possess an antipyretic effect on LPS induced fever [36] mainly through inhibition of COX2 and PGE2 release from POA [37]. On the other hand, there are some data opposing the general thermolytic action of NO suggested by Dantonio et al., (2016) and Perotti et al., (1999) proving that the NOS inhibitors like N^G-nitro-L-arginine methyl ester and 7-nitroindazole can attenuate LPS induced fever but do not affect LPS induced rise in PGE2 within the brain [38,39].

5.2. Hydrogen sulphide

Hydrogen sulphide (H_2S) is considered as an a typical gaseous neurotransmitter regulating several physiological functions like neuromodulation, vascular and smooth muscle relaxation. Predominantly, cystathionase, cystathionine beta synthase (CBS) and 3-Mercaptopyruvate sulphur transferase (MST) catalyzes the synthesis of H_2S from cysteine in mammalian tissues [40]. Even though H_2S has role in number of biological activities, little is known about its function in thermoregulation. Kwiatkoski et al., (2013) provide data with the notion that LPS can cause a reduction in the levels of H_2S in anteroventral pre-optic region of the hypothalamus (AVPO) and increases the AVPO COX-2/microsomal PGE synthase-1 (mPGES-1) activity. Moreover, it was observed that amino-oxyacetate (CBS inhibitor) potentiated LPS-induced PGE2 production in POA, where Na_2S (H_2S donor) has the opposite effect. It was also noted that Na_2S administration not only attenuated fever and PGE2 production but also inhibited LPS mediated reduction of cyclic AMP in AVPO [41].

5.3. Carbon monoxide

Carbon monoxide (CO) gas has long been considered as a toxic compound, owing to its ability to bind haemoglobin with higher affinity than oxygen. Still, there are evidences from researchers that endogenous CO, aby-product of inducible hemeoxygenase (HO-1) enzyme can modulate inflammation, inhibits LPS induced production of cytokines (both *in vivo* and *in vitro*), and consequently exhibits cyto-protective and anti-inflammatory functions beneficial for the resolution of acute inflammation. One of the novel CO releasing molecule tricarbonyl-dichlororuthenium (II) dimer (CORM-2) is considered as the most preferred molecule to investigate the role of CO in many of the biological functions. When CORM-2 was incorporated along with LPS in human umbilical vein endothelial cells (HUVEC), there observed down regulation of polymorpho nuclear cell adhesion, iNOS, ICAM-1 and NFκB expression followed by reduction in oxidative stress andinflammatory response to LPS stimulation [42]. *In vivo* result from Wang et al., (2016) was supportive to the above findings that CORM-2 significantly reduces IL-1β and Malondialdehyde (MDA) production in septic mice. It also helped to minimize tissue injury as well as neutrophil infiltration in vital organs like lungs and liver [43].

Immuno responsive gene 1 (IRG1) protein has role in endotoxin

tolerance by increasing the expression of A20 proteins in macrophages. A20 acts as a negative regulator of NFκB activation for the inflammasome mediated production of TNF-α. Functional link between HO-1 mediated CO production and IRG1 stimulation was correlated by Uddin et al., (2016) and was found that murine macrophages treated with CORM-2 (20 mM) showed upregulated expression of both HO-1 and IRG1 proteins in cells. In CORM-2 treated RAW264.7 cellstime- and dose-dependent rise in A20 protein level was notified. Rather than *in vitro* experiments, anti-inflammatory action of CO/HO-1 system was well demonstrated through an *in vivo* endotoximic mouse model. It was found that enhanced expression of IRG1 and A20 via CO/HO-1 system improved the anti-inflammatory response of the animal[44].

Similarly, it was noted that introduction of CORM-2, HO-1 inducers (hemin and cobalt protoporphyrin IX) and transfection of HO-1 inhibited chemotactic movement of immune cells, translocation of High-mobility group box 1 (HMGB1; a chromatin-binding protein that mediate various cellular responses) and release of pro-inflammatory cytokines in situations like severe sepsis. *In vitro* experiment with RAW264.7 macrophage cells treated with LPS (1 μg/ml) showed increased release of HMGB1, TNF-α, IL-1β and INF-β. The entire network of inflammatory reactions was reversed by the inhibition of HMGB1 in presence of HO-1 inducers and CORM-2. Delayed hemin and CORM-2 administration also protected mice from LPS (15 mg/kg) induced endotoxemia *in vivo* [45]. The effect of CO on molecular signalling pathway associated with LPS stimuli was further studied by Chicora et al., (2009) using human monocytic cell line THP-1. Cells were stimulated with LPS (1 μg/ml) and the anti-inflammatory effect of CO was assessed by investigating expression of genes responsible for inflammatory reactions, stress kinase pathway and NFκB signalling. It was observed that CO has inhibitory effect on upregulated genes associated with LPS interaction in THP1 cells. Genes which are upregulated after LPS challenge can initiate and propagate inflammation within the cells. CCL3, CXCL1, CXCL3, PTX3, PDE4B, NFKBIA, ATF3 and EGR family members other than EGR1, miR-155 and TNFAIP3 are the examples of CO responsive transcripts [46]. The overall interaction of antipyretic mediators in hypothalamus for resolution of pyrexia is depicted in Fig. 3.

6. Eicosanoids

In addition to PGE2, eicosanoids derived from other pathway of Arachidonic acid (AA) metabolism also have thermoregulatory role in inflammation and fever. Prostaglandin D2 (PGD2) has both anti- and pro-inflammatory roles at different stages of inflammation, and these opposing effects are mediated by distinct signalling mechanisms through interaction with D-prostanoid receptor (DP) orchemo attractant receptor-homologous molecule expressed on Th2 cells (CRTH2) [47,48]. Presence of 10 μM Prostaglandin J2 (PGJ2) nullified the consequence of LPS (0.1, 50, and 100 μg/ml) interaction with rat alveolar macrophage cells via reducing the production of NO, TNF-α and MIP-2. However, the magnitude of inhibition was time dependent and was greatest at 2 h before and least at 4 h after LPS addition. Since peroxisome proliferator-activated receptor-γ(PPAR-γ) was expressed in multiple tissues, the ligand specific activation of the PPAR-γ transcription factor has shown to inhibit pro-inflammatory gene expression. Porter et al., 2007 carried outRT-PCR analysis of PPAR-γ and NFκB p50 subunit mRNA expression in macrophages after LPS and PGJ2 treatment. Exposure of LPS to alveolar macrophages stimulated the production of NO metabolites, where PGJ2 significantly inhibited LPS-induced NO production when macrophages incubated with PGJ2 prior to LPS exposure. There observed time dependent inhibition of TNF-α and MIP-2 by PGJ2 where maximum inhibition was detected with addition of PGJ2 in macrophage culture 2 h before LPS. However, PGJ2 did not possess effect on production of TNF-α and MIP-2 when it was introduced 4 h after LPS. It was noted that PGJ2 had slight inhibitory role to LPS induced NF-κB p50 subunit mRNA expression where no such action was found against LPS mediated reduction of PPAR-γmRNA expression. Hence from the study it was concluded that the mechanism underlying PGJ2 inhibition was not through a PPAR-γ, but rather by NFκB dependent mechanism [49].

15-Deoxy-Δ^{12} and Δ^{14}-PGJ2 inhibited LPS induced RelA (v-rel avian reticuloendotheliosis viral oncogene homolog A) phosphorylation, $I_kB\alpha$

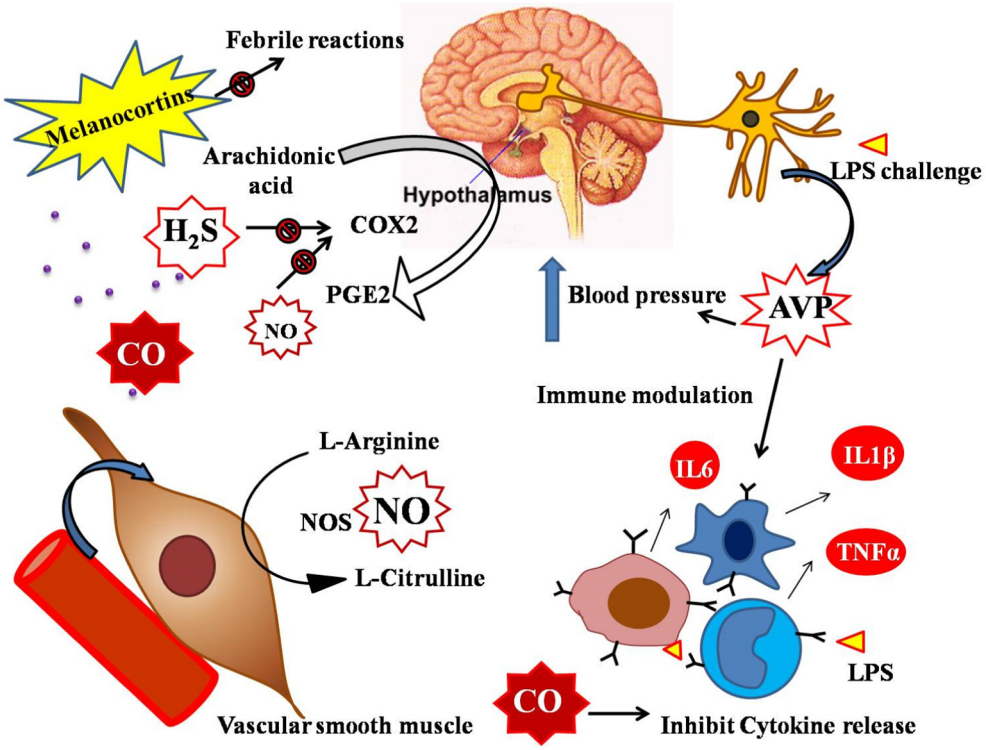

Fig. 3. Interaction of Antipyretic mediators in hypothalamus.

phosphorylation, ERK-signalling cascade and IL-6 gene expression through triggered protein phosphatise 2A activity in the colonic and intestinal epithelial cell line. The antagonistic effect of MEK1 Inhibitor like PD98059 on IL-6 expression articulates the fact that anti-inflammatory activity exhibited by PGJ2 was mediated via induction of MEK/ERK signalling cascade. However, 15-D-PGJ2 and high affinity PPAR-γ ligandrosiglitazone had no effect on LPS-induced degradation and gene expression of $I_{\kappa}B\alpha$ as well as nuclear translocation and DNA binding activity of RelA [50]. PGJ2 also showed inhibition of IL-1β, nitrite, COX-2 and iNOS expression in RAW264.7 and CD-1 mouse peritoneal macrophages after LPS and IFN-γ exposure [51].

Leukotriens (LTs) are another metabolite of AA released by the catalytic action of enzyme lipoxygenase (LOX). There are mainly three types of mammalian LOX such as 5, 12 and 15 LOX, out of which 5-LOX is the major enzyme that transforms AA to LTs. LTs are classified as LTB_4 and Cysteinyl LTs (LTC_4, LTD_4, and LTE_4) which has role in neutrophil chemotaxis and asthma through interaction with G protein coupled receptors expressed on inflammatory cells [52]. The formation of LTC_4 catalysed by LTC_4 synthase enzyme represents the first committed step in the production of cysteinyl LTs. RT-PCR analysis of LOX and LTC_4 synthase mRNA expression at rat hepatocytes and kupffer cells demonstrated that co-operate transcellular synthesis of one or more enzymes are necessary for LTs production within the liver. Production of these enzymes was increased after administration of LPS (3 mg/kg) and contributed toxic liver injury during inflammation reaction [53].

In contrast, Serio et al., (2003) reported the inhibitory effect of LPS on LTC4 synthase production within THP1cells through Fib dependent pathway. There obtained dose and time dependent inhibition of LTC4 synthase after LPS exposure [54]. Similarly, there are several controversies about the anapyretic activity of LTs and was discussed in detail by Kozak et al., (2004) in their review article. It discussed mostly about the role of non-prostaglandin eicosanoids and their inhibitors on temperature regulation primarily in mice and other lab animals [55]. Fig. 4 represents inhibitory effect of eicosanoids to the production of inflammatory cytokines.

7. Exogenous antipyretic drugs

NSAIDs are broad group of drugs having effects like reduction of fever, pain, and inflammation in common.While considering NSAIDs, their antipyretic activity is proposed to be related to their ability to inhibit synthesis and release of prostaglandins. Aspirin is the best example of antipyretic drug with well-known anti-inflammatory, analgesic and anti-platelet activities. Comparative study with aspirin and NOSH-aspirin (NBS-1120; a novel nitric oxide- and hydrogen sulphide-releasing hybrid) revealed the safer aspects of NOSH-aspirin with slight reduction in production of pro inflammatory cytokine TNF-α in Wistar rats compared to which induced by aspirin. Similar to aspirin NOSH-aspirin caused reduction in PGE2, where alternate result was found with production of MDA and superoxide dismutase. NOSH-aspirin reduced the MDA level and increased superoxide dismutase activity in stomach and paw exudates where aspirin has opposite effect [56]. Both aspirin and NOSH-aspirin showed equally effective antipyretic activity via reducing body temperature and mechanical pain in rats after LPS administration. Although NOSH-aspirin was found to be a safer alternate to aspirin with less index of lipid peroxidation, gastric ulcerative damage and high index of chemo preventive nature has also found to be in prevalence [56]. Likewise, NOSH-sulindac composition showed novel analgesic, anti-inflammatory, anti-cancerous and antipyretic activities compared to sulindac alone [57].

Hinz et al., (2008) presented a data regarding the action of most common antipyretic drug acetaminophen which contributes its antipyretic effect through inhibition of COX2 enzymes in humans (*in vivo*) and monocytes (*in vitro*) isolated from human blood [58]. It was observed that acetaminophen (100 μmol/L) inhibited the production of PGE2 in human whole blood and COX2 enzyme activity in monocyte culture [58]. Treatment with medicinal plant extract was also used as a source of alternative tool for traditional antipyretic therapy. Extracts taken from plants like *Eleucineindica* [59], *Amaranthusspinosus* [60] and

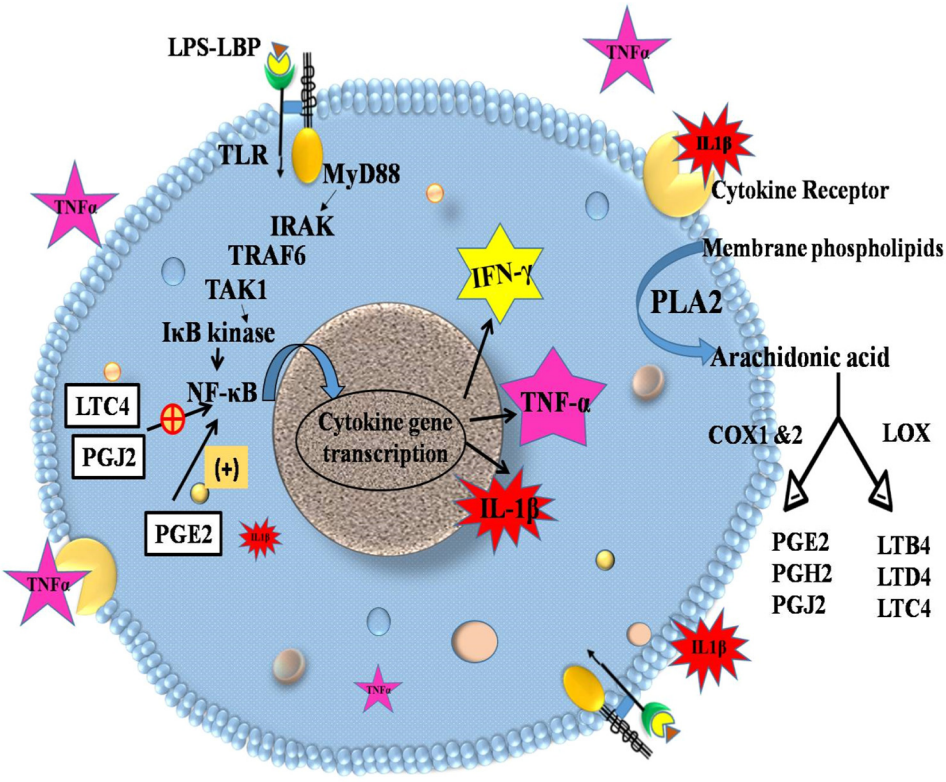

Fig. 4. Inhibitory effect of Eicosanoids on release of endogenous pyrogens.

Pothosscandens L [61] showed strong antioxidant activity *in vitro* and antipyretic activity *in vivo* in albino rats after subcutaneous administration of Brewer's yeast suspension as exogenous pyrogen.

Apart from NSAIDs, some narcotic drugs like morphine also exhibit antipyretic activity. Inhibition of TLR9 mediated NFκB signalling by morphine within the mice macrophages after *Streptococcus pneumonia* infection was informative about the potential antipyretic activity of the same in *in vivo* system [62]. Since NFκB signalling is necessary for the release of pyretic cytokines like IL-6, IL-1β and TNF-α, action of morphine should reduce endogenous pyrogens mediated fever. Recently it was reported in a clinical study by Mendieta *et al.,* (2014) that administration of morphine (i.v, 0.05 mg/kg/h) successfully eliminated fever in patient who was suffering from consistent fever even after administration of acetaminophen and metamizole for about 31 days [63].

8. Conclusion

Fever; one of the central expressions of immune response remains as a fascinating problem for both clinicians and basic scientists because of the fortuitous defects in regulation of immune system and hence can lead to severe illness and inflammatory diseases. Fever induced by exogenous pyrogens and associated production of endogenous pyrogens from phagocytic cells especially monocytes and macrophages initiate upward shift in temperature, initially begins at the periphery of the body. The released inflammatory mediators convey the alarming signals to the immune cells for further actions and also transmit messages to thermoregulatory centre of brain to generate defence responses appropriate to eliminating the offensive agents from body to restoring good health.

When the inflammatory stimulus is cleared, endogenous pyrogens production ceases and set point reverts back to normal by several thermoregulatory mechanisms of the body. The important antipyretic mechanisms exhibited by the immune system include: production of antipyretic cytokines like IL-10, IL-1 receptor antagonists, antipyretic neuropeptides (ACTH, α-MSH), eicosanoids (PGD2, PGJ2, LTs), gaseous neurotransmitters (NO, CO, H_2S *etc*.,) and hormones (arginine vasopressin, glucocorticoids). If the immune system fails to restore homeostasis, then different types of NSAIDs should be used as exogenous antipyretic drugs. The present review is meant to discuss the importance of knowing the mechanism of antipyresis to improve the efficacy of antipyretic therapeutics and their controlled usage to set a healthy diagnosis and treatment strategy.

Conflict of interest

The authors declare that they have no conflict of interest.

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References

[1] C.A. Wunderlich, J.C. Reeve, The course of the temperature in diseases: a guide to clinical thermometry, Am. J. Med. Sci. 57 (1869) 425–447.
[2] J. Roth, C.M. Blatteis, Mechanisms of fever production and lysis: lessons from experimental LPS fever, Compr. Physiol. 4 (4) (2011) 1563–1604.
[3] P.L. McCarthy, Down-regulation of cytokine action, Baillieres Clin. Haematol. 7 (1) (1994) 153–177.
[4] Rothwell NJ. Cytokines in the nervous system. Springer Science & Business Media; Ist edition.1996; 73-86.
[5] S.M. Opal, V.A. De Palo, Anti-inflammatory cytokines, Chest. 117 (4) (2000) 1162–1172.
[6] M. Sawada, A. Suzumura, H. Hosoya, T. Marunouchi, T. Nagatsu, Interleukin-10 inhibits both production of cytokines and expression of cytokine receptors in microglia, J. Neurochem. 72 (4) (1999) 1466–1471.
[7] T. Cartmell, C. Ball, A.F. Bristow, D. Mitchell, S. Poole, Endogenous interleukin-10 is required for the defervescence of fever evoked by local lipopolysaccharide induced and Staphylococcus aureus induced inflammation in rats, J. physiol. 549 (2) (2003) 653–664.
[8] L.R. Leon, W. Kozak, K. Rudolph, M.J. Kluger, An antipyretic role for interleukin-10 in LPS fever in mice, Am. J. Physiol. Regul. Integr. Comp. Physiol. 276 (1) (1999) 81–89.
[9] L.M. Harden, C. Rummel, G.N. Luheshi, S. Poole, R. Gerstberger, J. Roth, Interleukin-10 modulates the synthesis of inflammatory mediators in the sensory circumventricular organs: implications for the regulation of fever and sickness behaviors, J. Neuroinflam. 10 (1) (2013) 790.
[10] L. Crepaldi, L. Silveri, F. Calzetti, C. Pinardi, M.A. Cassatella, Molecular basis of the synergistic production of IL-1 receptor antagonist by human neutrophils stimulated with IL-4 and IL-10, Int. Immunol. 14 (10) (2002) 1145–1153.
[11] W.E. Ip, N. Hoshi, D.S. Shouval, S. Snapper, R. Medzhitov, Anti-inflammatory effect of IL-10 mediated by metabolic reprogramming of macrophages, Science 356 (6337) (2017) 513–519.
[12] D.W. Cain, J.A. Cidlowski, Immune regulation by glucocorticoids, Nat. Rev. Immunol. 17 (4) (2017) 233–247.
[13] R.N. Soriano, L.G. Branco, Reduced stress fever is accompanied by increased glucocorticoids and reduced PGE2 in adult rats exposed to endotoxin as neonates, J. Neuroimmunol. 225 (1–2) (2010) 77–81.
[14] R. Cabrera, S.M. Korte, E.G. Lentjes, F. Romijn, E. Schönbaum, A. De Nicola, E.R. De Kloet, The amount of free corticosterone is increased during lipopolysaccharide-induced fever, Life Sci. 66 (7) (2000) 553–562.
[15] C. Parent, H.B. Nguyen, X. Wen, J. Diorio, M.J. Meaney, T.Y. Zhang, Maternal care modulates the febrile response to lipopolysaccharide through differences in glucocorticoid receptor sensitivity in the rat, Brain Behav Immun. 65 (2017) 239–250.
[16] R.N. Soriano, L.G. Nicoli, E.C. Carnio, L.G. Branco, Exogenous ghrelin attenuates endotoxin fever in rats, Peptides 32 (11) (2011) 2372–2376.
[17] L. Wang, N.R. Basa, A. Shaikh, A. Luckey, D. Heber, D.H. St-Pierre, Y. Taché, LPS inhibits fasted plasma ghrelin levels in rats: role of IL-1 and PGs and functional implications, Am. J. Physiol. Gastrointest. Liver Physiol. 291 (4) (2006) 611–620.
[18] T. Waseem, M. Duxbury, H. Ito, S.W. Ashley, M.K. Robinson, Exogenous ghrelin modulates release of pro-inflammatory and anti-inflammatory cytokines in LPS-stimulated macrophages through distinct signaling pathways, Surgery 143 (3) (2008) 334–342.
[19] A.L. Beynon, M.R. Brown, R. Wright, M.I. Rees, I.M. Sheldon, J.S. Davies, Ghrelin inhibits LPS-induced release of IL-6 from mouse dopaminergic neurones, J. Neuroinflam. 10 (1) (2013) 811.
[20] J. Song, S.M. Kang, K.M. Lee, J.E. Lee, The protective effect of melatonin on neural stem cell against LPS-induced inflammation, Bio Med Res Int. 2015 (2015) 1–13.
[21] G.M. Yu, H. Kubota, M. Okita, T. Maeda, The anti-inflammatory and antioxidant effects of melatonin on LPS-stimulated bovine mammary epithelial cells, PloS one 12 (5) (2017) 1–17.
[22] E. Tyagi, R. Agrawal, C. Nath, R. Shukla, Effect of melatonin on neuroinflammation and acetylcholine esterase activity induced by LPS in rat brain, Eur. J. Pharmacol. 640 (1–3) (2010) 206–210.
[23] H. Wang, D.X. Xu, J.W. Lv, H. Ning, W. Wei, Melatonin attenuates lipopolysaccharide (LPS)-induced apoptotic liver damage in D-galactosamine-sensitized mice, Toxicology 237 (1–3) (2007) 49–57.
[24] I. Gantz, T.M. Fong, The melanocortin system, Am. J. Physiol. Endocrinol. Metab. 284 (3) (2003) 468–474.
[25] J.B. Tatro, Endogenous antipyretics, Clin. Infect. Dis. 31 (2000) 190–201.
[26] W.G. Clark, M.I. Holdeman, J.M. Lipton, Analysis of the antipyretic action of alpha melanocyte stimulating hormone in rabbits, J Physiol. 359 (1) (1985) 459–465.
[27] Q.H. Huang, M.L. Entwistle, J.D. Alvaro, R.S. Duman, V.J. Hruby, J.B. Tatro, Antipyretic role of endogenous melanocortins mediated by central melanocortin receptors during endotoxin-induced fever, J. Neurosci. 17 (9) (1997) 3343–3351.
[28] W.H. Sawyer, R.A. Munsick, H.B. Van Dyke, Antidiuretic hormones, Circulation 21 (5) (1960) 1027–1037.
[29] T.C. Peng, C.J. Huang, Vasopressin inhibits endotoxin-induced upregulation of inflammatory mediators in activated macrophages, Tzu Chi Med. J. 25 (3) (2013) 150–154.
[30] J. Park, E.Y. Eo, K.H. Lee, J.S. Park, J.H. Lee, C.G. Yoo, C.T. Lee, Y.J. Cho, The anti-inflammatory effect of arginine-vasopressin on lipopolysaccharide-induced IκBα/nuclear factor-κB cascade, Korean J. Crit. Care Med. 30 (3) (2015) 151–157.
[31] K. Palin, M.L. Moreau, J. Sauvant, H. Orcel, A. Nadjar, A. Duvoid-Guillou, J. Dudit, A. Rabié, F. Moos, Interleukin-6 activates arginine vasopressin neurons in the supraoptic nucleus during immune challenge in rats, Am. J. Physiol. Endocrinol. Metab. 296 (6) (2009) 1289–1299.
[32] F. Nava, G. Carta, L.W. Haynes, Lipopolysaccharide increases arginine-vasopressin release from rat suprachiasmatic nucleus slice cultures, Neurosci. Lett. 288 (3) (2000) 228–230.
[33] F. Shimizu, T. Kasai, A. Takamata, Involvement of central angiotensin II type 1 receptors in LPS-induced systemic vasopressin release and blood pressure regulation in rats, J. Appl. Physiol. 106 (6) (2009) 1943–1948.
[34] I.M. Qahwash, C.A. Cassar, R.P. Radcliff, G.W. Smith, Bacterial lipopolysaccharide-induced coordinate downregulation of arginine vasopressin receptor V3 and corticotropin-releasing factor receptor 1 messenger ribonucleic acids in the anterior pituitary of endotoxemic steers, Endocrine 18 (1) (2002) 13–20.
[35] A.A. Steiner, L.G. Branco, Nitric oxide in the regulation of body temperature and fever, J. Therm. Biol. 26 (4–5) (2001) 325–330.
[36] A.V. Gourine, Pharmacological evidence that nitric oxide can act as an endogenous

antipyretic factor in endotoxin-induced fever in rabbits, Gen. Pharmacol. 26 (4) (1995) 835–841.
[37] C. Feleder, V. Perlik, C.M. Blatteis, Preoptic nitric oxide attenuates endotoxic fever in guinea pigs by inhibiting the POA release of norepinephrine, Am. J. Physiol. Regul. Integr. Comp. Physiol. 293 (3) (2007) 1144–1151.
[38] V. Dantonio, M.E. Batalhão, M.H. Fernandes, E.N. Komegae, G.A. Buqui, N.P. Lopes, L.H. Gargaglioni, É.C. Carnio, A.A. Steiner, Bícego KC. Nitric oxide and fever: immune-to-brain signaling vs. thermogenesis in chicks, Am. J. Physiol. Regul. Integr. Comp. Physiol. 310 (10) (2016) 896–905.
[39] C.A. Perotti, M.S. Nogueira, J. Antunes-Rodrigues, E.C. Carnio, Effects of a neuronal nitric oxide synthase inhibitor on lipopolysaccharide-induced fever, Braz. J. Medbiol. Res. 32 (11) (1999) 1381–1387.
[40] P. Kamoun, Endogenous production of hydrogen sulfide in mammals, Amino acids. 26 (3) (2004) 243–254.
[41] M. Kwiatkoski, R.N. Soriano, R.M. Araujo, L.U. Azevedo, M.E. Batalhao, H.D. Francescato, T.M. Coimbra, E.C. Carnio, L.G. Branco, Hydrogen sulfide inhibits preoptic prostaglandin E2 production during endotoxemia, Exp. Neurol. 240 (2013) 88–95.
[42] B. Sun, X. Zou, Y. Chen, P. Zhang, G. Shi, Preconditioning of carbon monoxide releasing molecule-derived CO attenuates LPS-induced activation of HUVEC, Int. J. Biol. Sci. 4 (5) (2008) 270–278.
[43] X. Wang, W. Qin, M. Song, Y. Zhang, B. Sun, Exogenous carbon monoxide inhibits neutrophil infiltration in LPS-induced sepsis by interfering with FPR1 via p38 MAPK but not GRK2, Oncotarget 7 (23) (2016) 34250–34265.
[44] M.J. Uddin, Y. Joe, S.K. Kim, S.O. Jeong, S.W. Ryter, H.O. Pae, H.T. Chung, IRG1 induced by heme oxygenase-1/carbon monoxide inhibits LPS-mediated sepsis and pro-inflammatory cytokine production, Cell Mol. Immunol. 13 (2) (2016) 170–179.
[45] K. Tsoyi, T.Y. Lee, Y.S. Lee, H.J. Kim, H.G. Seo, J.H. Lee, K.C. Chang, Heme-oxygenase-1 induction and carbon monoxide-releasing molecule inhibit lipopolysaccharide (LPS)-induced high-mobility group box 1 release in vitro and improve survival of mice in LPS-and cecal ligation and puncture-induced sepsis model in vivo, Mol. Pharmacol. 76 (1) (2009) 173–182.
[46] M. Chhikara, S. Wang, S.J. Kern, G.A. Ferreyra, J.J. Barb, P.J. Munson, R.L. Danner, Carbon monoxide blocks lipopolysaccharide-induced gene expression by interfering with proximal TLR4 to NF-κB signal transduction in human monocytes, PloS one 4 (12) (2009) 8139.
[47] T. Murata, K. Aritake, Y. Tsubosaka, T. Maruyama, T. Nakagawa, M. Hori, H. Hirai, M. Nakamura, S. Narumiya, Y. Urade, H. Ozaki, Anti-inflammatory role of PGD2 in acute lung inflammation and therapeutic application of its signal enhancement, Proc. Natl. Acad. Sci. U S A. 110 (13) (2013) 5205–5210.
[48] M. Ishii, K. Asano, H. Namkoong, S. Tasaka, K. Mizoguchi, T. Asami, H. Kamata, Y. Kimizuka, H. Fujiwara, Y. Funatsu, S. Kagawa, CRTH2 is a critical regulator of neutrophil migration and resistance to polymicrobial sepsis, J. Immunol. 188 (2012) 5655–5664.
[49] D.W. Porter, M. Wolfarth, S.H. Young, M.K. Rao, T. Meighan, M. Barger, M.E. Andrew, L.J. Huffman, PGJ2 inhibition of LPS-induced inflammatory mediator expression from rat alveolar macrophages, J. Toxicol. Environ. Health Part A 70 (23) (2007) 1967–1976.
[50] P.A. Ruiz, S.C. Kim, R.B. Sartor, D. Haller, 15-Deoxy-Δ12, 14-prostaglandin J2-mediated ERK signaling inhibits gram-negative bacteria-induced RelA phosphorylation and interleukin-6 gene expression in intestinal epithelial cells through modulation of protein phosphatase 2A activity, J. Biol. Chem. 279 (34) (2004) 36103–36111.
[51] L.B. Maggi, H. Sadeghi, C. Weigand, A.L. Scarim, M.R. Heitmeier, J.A. Corbett, Anti-inflammatory actions of 15-deoxy-delta 12, 14-prostaglandin J2 and troglitazone: evidence for heat shock-dependent and-independent inhibition of cytokine-induced inducible nitric oxide synthase expression, Diabetes 49 (3) (2000) 346–355.
[52] J.N. Sharma, L.A. Mohammed, The role of leukotrienes in the pathophysiology of inflammatory disorders: is there a case for revisiting leukotrienes as therapeutic targets? Inflammo. Pharmacology. 14 (1–2) (2006) 10–16.
[53] K. Shimada, J. Navarro, D.E. Goeger, S.B. Mustafa, P.H. Weigel, S.A. Weinman, Expression and regulation of leukotriene synthesis enzymes in rat liver cells, Hepatology 28 (5) (1998) 1275–1281.
[54] K.J. Serio, S.C. Johns, L. Luo, C.R. Hodulik, T.D. Bigby, Lipopolysaccharide down-regulates the leukotriene C4 synthase gene in the monocyte-like cell line, THP-1, J. Immunol. 170 (4) (2003) 2121–2128.
[55] W. Kozak, V. Fraifeld, Non-prostaglandin eicosanoids in fever and anapyrexia, Front Biosci. 9 (2004) 3339–3355.
[56] R. Kodela, M. Chattopadhyay, C.A. Velázquez-Martínez, K. Kashfi, NOSH-aspirin (NBS-1120), a novel nitric oxide-and hydrogen sulfide-releasing hybrid has enhanced chemo-preventive properties compared to aspirin, is gastrointestinal safe with all the classic therapeutic indications, Biochem Pharmacol. 98 (4) (2015) 564–572.
[57] K. Kashfi, M. Chattopadhyay, R. Kodela, NOSH-sulindac (AVT-18A) is a novel nitric oxide-and hydrogen sulfide-releasing hybrid that is gastrointestinal safe and has potent anti-inflammatory, analgesic, antipyretic, anti-platelet, and anti-cancer properties, Redox biol. 6 (2015) 287–296.
[58] B. Hinz, O. Cheremina, K. Brune, Acetaminophen (paracetamol) is a selective cyclooxygenase-2 inhibitor in man, FASEB J. 22 (2) (2008) 383–390.
[59] E.O. Ettebong, A.N. Paul, Antipyretic and antioxidant activities of Eleucineindica, J Phytopharmacol. 4 (4) (2015) 235–242.
[60] B.S. Kumar, K. Lakshman, K.N. Jayaveera, D.S. Shekar, A.A. Kumar, B. Manoj, Antioxidant and antipyretic properties of methanolic extract of Amaranthusspinosusleaves, APJTM 3 (9) (2010) 702–706.
[61] T. Sajeesh, K. Arunachalam, T. Parimelazhagan, Antioxidant and antipyretic studies on Pothosscandens L, APJTM 4 (11) (2011) 889–899.
[62] J. Wang, R.A. Barke, R. Charboneau, R. Schwendener, S. Roy, Morphine induces defects in early response of alveolar macrophages to Streptococcus pneumoniae by modulating TLR9-NF-κB signaling, J Immunol. 180 (5) (2008) 3594–3600.
[63] H.Z. Mendieta, J.G. Arriaga, Remission of central fever with morphine post traumatic brain injury, Le J. Med. libanais. LMJ. 62 (1) (2014) 57–61.