Medical Hypotheses 73 (2009) 470–472

Contents lists available at ScienceDirect

Medical Hypotheses

journal homepage: www.elsevier.com/locate/mehy

Acupuncture: A novel hypothesis for the involvement of purinergic signalling

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ARTICLE INFO

Article history:
Received 14 May 2009
Accepted 15 May 2009

SUMMARY

The hypothesis is summarised schematically in Fig. 1.

It is proposed that mechanical deformation of the skin by needles and application of heat or electrical current leads to release of large amounts of ATP from keratinocytes, fibroblasts and other cells in skin; the ATP then occupies specific receptor subtypes expressed on sensory nerve endings in the skin and tongue; the sensory nerves send impulses through ganglia to the spinal cord, the brain stem, hypothalamus and higher centres; the brain stem and hypothalamus contain neurons that control autonomic functions, including cardiovascular, gastrointestinal, respiratory, urinogenital and musculo-skeletal activity. Impulses generated in sensory fibres in the skin connect with interneurons to modulate (either inhibition or facilitation) the activities of the motoneurons in the brain stem and hypothalamus to change autonomic functions; specifically activated sensory nerves, via interneurons, also inhibit the neural pathways to the pain centres in the cortex.

Background to purinergic signalling

ATP has been well established as an intracellular energy source for many years. However, in 1972 the concept of purinergic signalling was introduced, proposing that ATP also acts as an extracellular signalling molecule [1]. This concept was rejected by many for the next 20 years, but when the receptors for ATP and its breakdown product adenosine were cloned and characterised in the early 1990s, the concept was accepted and purinergic signalling is now a rapidly expanding field (see [2]). Purinergic-related drugs are being developed to treat a variety of diseases. For example, clopidogrel, an antagonist to the G protein-coupled receptor subtype on platelets that mediates aggregation, is a widely used drug against stroke and thrombosis (it made US$8.6 billion in 2007). Clinical trials for other purinergic agents are in progress for bladder incontinence, dry eye, cystic fibrosis, osteoporosis, pain and cancer (see Fig. 1).

Supporting evidence

ATP release

While large amounts of ATP are released from damaged or dying cells, it has become clear that ATP transport from many cells in response to mechanical deformation, hypoxia, heat and electrical currents is a physiological event, which occurs without damage to the cells. For example, changes in blood flow results in shear stress releasing ATP from endothelial cells leading to vasodilation via nitric oxide [3], and ATP is released from urothelial cells in the bladder and ureter leading to stimulation of suburothelial sensory nerves and from epithelial cells of the airways [3]. There is evidence for release of ATP from keratinocytes in response to mechanical stimulation [4,5] as well as fibroblasts [6] and immune cells [7].

ATP receptors on sensory neurons

Implicit in purinergic signalling is the presence of specific receptors for purines. Two families of purine receptors were recognised in 1978, P1 receptors for adenosine and P2 receptors for ATP. Two families of P2 receptors were proposed in 1985 and the molecular structure and second messenger agents involved discovered in the 1990s. Seven P2X ligand-gated ion channel receptor subtypes and eight P2Y G protein-coupled receptor subtypes are currently established (see [2]). $P2X_3$ homomultimer and $P2X_{2/3}$ heteromultimer receptors were cloned in 1995 and shown to be located almost exclusively on sensory nerve endings [8].

These receptors have been shown with immunohistochemistry to be located on nerve endings in the skin and are particularly abundant in the tongue also used for acupuncture [9] (Fig. 2). Electrical recording from an isolated tongue-nerve preparation showed increased activity in the lingual nerves supplying the tongue during mechanical stimulation of the tongue that was mimicked by ATP and attenuated with $P2X_3$ receptor antagonists [10]. Similarly, distension of the ureter led to release of substantial amounts of ATP and evoked a discharge in the suburothelial sensory nerves that was mimicked by ATP and reduced by 2′,3′-O-(2,4,6-trinitrophenyl)-ATP, a potent $P2X_3$ and $P2X_{2/3}$ receptor antagonist (see [2]).

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doi:10.1016/j.mehy.2009.05.031

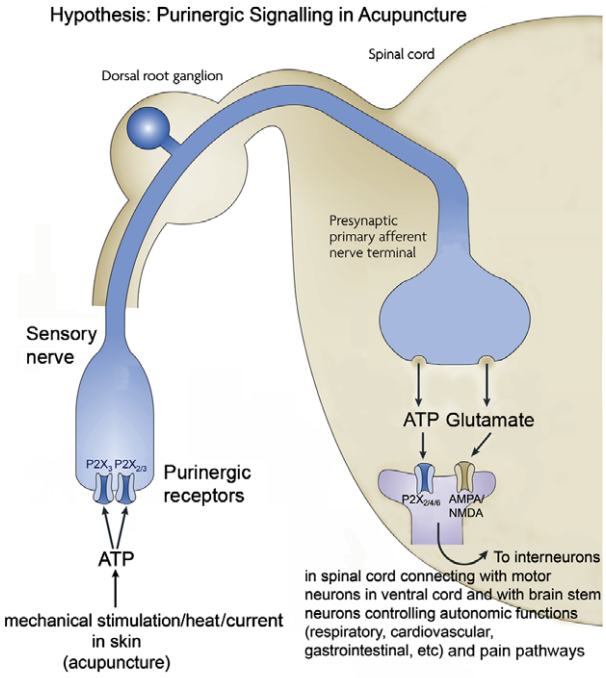

Fig. 1. Schematic hypothesis of purinergic signalling in acupuncture – see summary for explanation.

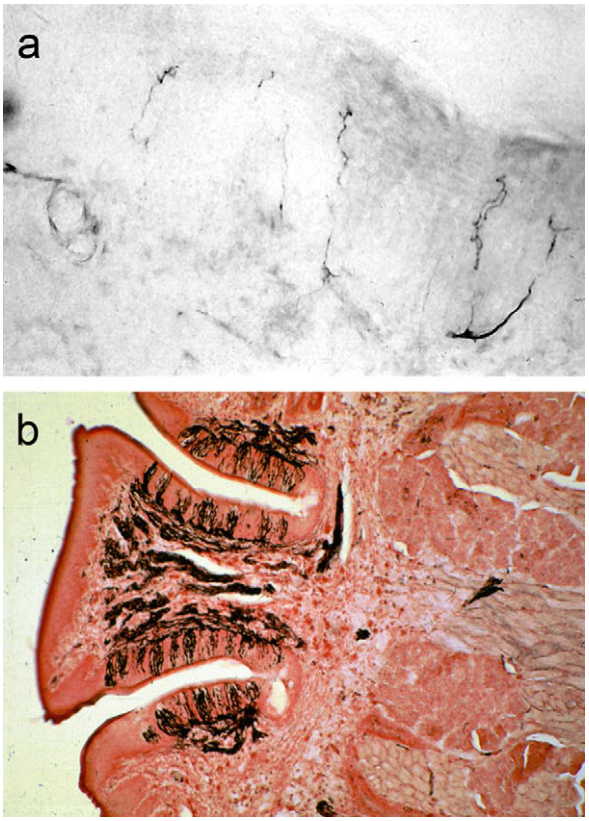

Fig. 2. Sensory nerve fibres showing immunolocalisation of $P2X_3$ receptor in (a) skin and (b) tongue (courtesy of A. Alavi and G. Burnstock).

Sensory pathways to the brain centres controlling autonomic function and recognising pain

The sensory nerves supplying the body skin originate in dorsal root ganglia and their central connections are in lamina 2 of the dorsal spinal cord, while those supplying the tongue and skin of the head originate in the trigeminal ganglion. Then interneurons mediate modulatory pathways to the brain stem and hypothalamus, which are the control centres for autonomic functions, including respiratory, cardiovascular, urinogenital and gastrointestinal systems (see [11]).

It is suggested that sensory nerve activity initiated in the skin by acupuncture will have an inhibitory modulating effect on the spinoparabrachial and spinothalamic tracts to the brain pain centres, possibly via the release of endorphins in the thalamus and periaqueductal gray (see [12,13]).

Much is now known about the pre- and post-synaptic modulatory mechanisms that operate in the central nervous system and it is here that it is suggested that the sensory information involved in specific skin areas by acupuncture leads to modulation of different autonomic functions that are well established in the acupuncture literature (see [14]).

Concluding remarks

The proposed hypothesis for a mechanism underlying acupuncture has the advantage that tools are available so that every step can be tested experimentally. For example, very sensitive ATP assay techniques are now available (see [15]). Selective $P2X_3$ and $P2X_{2/3}$ receptor antagonists are available [16] and ecto-nucleotidase inhibitors have been identified to see if their application would enhance the effect of acupuncture by making more ATP available. It seems likely from experiments in both the bladder and intestine that ATP-sensitive low threshold sensory fibres mediate physiological events, while high threshold fibres mediate nociception [2,17]. However, increasing epidermal keratinocyte ATP release by stimulation of sodium channels, results in excessive activation of P2X receptors on nociceptive primary sensory axons [18]. This will need to be resolved for the sensory nerves supplying the skin and tongue, before approaches to enhancing the ATP-related responses to acupuncture are carried out, in case it results in pain. Free nerve endings of both A-δ and C classes of nerve fibres are located in the dermis and epidermis extending to the stratum granulosum and are activated by ATP [19].

Conflict of interest statement

The author has no competing interests that might be perceived to influence the results and discussion reported in this paper.

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