Evidence based formulation proves to exquisitely relieve pain and inflammation when applied topically

Observations made on the pain-relieving capacity of Polyactil on sports lesions in professional soccer players led to the development of Bioactil, the leading formulation for topical pain relief and of inflammation.

During a study done in a professional soccer team of Costa Rica a decade ago, athletes reported intense relief of pain and inflammation when Polyactil was applied to the lesion and then cooling the lesion with ice kept on the affected area for some 10 mins. This pain relief was not well understood at that moment. However, it was clear that Polyactil could resolve these closed lesions more rapidly than known methods adopted for many decades and, for the intervention group treated with Polyactil, leaving no fibrotic sequelae at ultrasound examination.

One of the positive control groups was treated with inhibitors of cyclooxigenase-2 or COX-2. This group did not have the same favorable outcome as the Polyactil group did. Polyactil became the predecessor upon which Bioactil was developed.

Since then the resolution phase of inflammation has been uncovered and studied extensively and it has become clear that for damaged tissues to produce Lipoxins and Resolvins, which pro-actively promote resolution of inflammation and possibly regenerative healing, the inducible COX-2 enzyme is essential. Resolution is a local process at the site of injury occurring via transcellular sequential biosynthesis which generates autocrine and paracrine lipid mediators from arachidonic acid released by action of lipoxygenases on the plasma membrane and subsequently metabolized by the COXs isoforms. Downstream in the sequence, it switches strategically at some point to generation of lipoxins from arachidonic acid and resolvins from essential polyunsaturated fatty acids, rather than prostaglandings and leukotrienes as in the preceding inflammatory phase (Samuelsson, Dahlén, Lindgren, Rouzer, & Serhan, 1987).

Although inhibitors of COX-2 are still used in clinical practice they are held now responsible for disrupting endogenous resolution mechanisms (Serhan, Chiang, & Van Dyke, 2008). For the above described switch to occur it has been shown that a second paradoxical peak of COX-2 expression, 350% more intensive than the one during the

inflammatory phase, sub-enters and coinciding fully with the production of resolution mediators (Gilroy et al., 1999).

Other researchers have shown that COX-2 inhibitors reduce the production of key local lipid mediators, leading to deficits in inflammation resolution (Fukunaga, Kohli, Bonnans, Fredenburgh, & Levy, 2005)(Willoughby, Moore, Colville-Nash, & Gilroy, 2000). It was found in an early study using a rat inflammation model (pleurisy) that the COX-2 inhibitor NS-398 reduced inflammation at 2 hours but enhanced inflammation at 48 hours. Therefore it is well proven that COX-2 essentially contributes to resolving inflammation and that its inhibition is a harmful intervention and in the longer term it is pronociceptive.

Pain from inflammation is induced by inflammatory mediators released after tissue insults and subsequent hypoxic injury microenvironments that follow. Nociceptor terminals (bidirectional signaling units) express receptors for all inflammatory mediators. Activation of these receptors causes hyperactivity of key transduction molecules, such as transient receptor potential subtype V1 (TRPV1) and A1(TRPA1) and conduction molecules such as sodium channels Nav1.7/1.8/1.9. As a result, the sensitivity and excitability of nociceptors are increased, via activation of protein kinases, such as protein kinase A (PKA), protein kinase C (PKC), and mitogen-activated protein kinases (MAPKs). This can be called a peripheral sensitization of pain (excitation thresholds drops). It is generally believed that inflammatory pain is driven by peripheral sensitization within primary sensory neurons in the dorsal root ganglion (DRG) (Hucho & Levine, 2007)(Basbaum, Bautista, Scherrer, & Julius, 2009).

Tissue injury-triggered hyperactivity of nociceptors will lead to increased release of neurotransmitters (e.g., glutamate) and neuromodulators [eg. substance P, enkephalins; neurokinin; serotonin and brain-derived neurotrophic factor (BDNF)] from nociceptor central terminals in the spinal cord, causing hyperactivity of postsynaptic dorsal horn neurons and sensitization. Neurotransmission during prolonged nociceptor excitation reaches beyond the DRGs and is able of establishing postsynaptic sensitization escalating pain to a more advanced and complex form of central sensitization (Ji, Kohno, Moore, & Woolf, 2003).

Activation of the N-methyl-D-aspartate receptor NMDAR plays an essential role in the induction and maintenance of central sensitization also called spinal cord synaptic plasticity (Latremoliere & Woolf, 2009)(Dubner & Ruda, 1992). Tissue injuryinduced spinal cord synaptic plasticity also is important for maintaining persistent pain and generating secondary pain outside the initial injury site (Ji et al., 2003)(Dubner & Ruda, 1992) Central sensitization contributes importantly to the development and maintenance of inflammatory pain (Ji et al., 2003)(Kuner, 2010). Long-term potentiation (LTP) in the spinal cord (Ruscheweyh, Wilder-Smith, Drdla, Liu, & Sandkühler, 2011) is a unique form of central sensitization for persistent pain development. The complexity of neurotransmission and sensitization of inflammatory pain suggests that a reductionist intervention directed to

inhibit one or two of the transmitters involved, either peripherally or centrally, is destined to fail.

Other studies have established that resolution mediators had a profound analgesic effect on the pain caused by inflammation and spinal cord synaptic plasticity in mice by inhibition of transient receptor potential channels TRPs in the nm range. Members of the Resolvin family inhibited TRPs as follows: RvD2 was found to be a remarkably potent inhibitor of TRPV1 (IC_{50} = 0.1 nm) and TRPA1 (IC_{50} = 2 nm) in primary sensory neurons, whereas RvE1 and RvD1 selectively inhibited TRPV1 (IC_{50} = 1 nm) and TRPA1 (IC_{50} = 9 nm), respectively (Park et al., 2011)(Morales-Lázaro, Simon, & Rosenbaum, 2013).

Transient receptor potential vanilloid 1 (TRPV1) and ankyryn 1 (TRPA1) are two critical types of TRP channels that are strongly implicated in the genesis of inflammatory pain (Caterina, 2000)(McMahon & Wood, 2006). Activation of TRPV1 and TRPA1 can enhance inflammatory pain not only via well demonstrated peripheral sensitization (Ji, Samad, Jin, Schmoll, & Woolf, 2002)(Bautista et al., 2006)(Dai et al., 2007) but also via central sensitization, by increasing glutamate release from primary afferent terminals to enhance synaptic transmission (Kosugi, Nakatsuka, Fujita, Kuroda, & Kumamoto, 2007).

So, the experimental success in reducing inflammatory pain is appealing and illustrative though the detailed signaling mechanisms of RvD2 in pain relief remain unclear. Therefore, inflammatory pain relief by topical application of Bioactil becomes very enlightening since it is resolution-driven and by correlation quite well defined as antagonistic to TRPV1 and TRPA1. Thus, the marked relief of pain that can be observed while inflammation is gradually reduced by Bioactil can be equated to the action if RvD2; not quite dissimilar in both facets of inflammation resolution and relief of inflammatory pain.

Bioactil's potentiation by application of cold which enhances its analgesic effect is even more revealing. Six of the 28 TRP channels from the three distinct TRP family subtypes are activated by temperature (TRPV1–4, TRPM8 and TRPA1), and are expressed in sensory neurons or in skin keratinocyte cells, which are peripheral targets of these nerves (Dhaka, Viswanath, & Patapoutian, 2006). At the same time, clinical evidence suggests that cold does enhance the resolution effect of Bioactil since if used together with cold, inflammation subsides sooner. The latter fact confirms how complex is the physiology of the sensory system and the pathophysiology of pain. But it must be conceded for this purpose that an agonistic effect is being exerted on TRPM8 by cold and not an inhibitory one. Indeed, most cold-sensitive neurons respond to menthol and display a thermal activation threshold of ∼ 25°C. TRPM8 is a cold and menthol-sensitive channel whose physiological characteristics match those of native cold currents and TRPM8-deficient mice show a very substantial loss of menthol and cold-evoked responses at the cellular or nerve fiber level. Likewise, these animals display severe deficits in cold-evoked behavioral responses (Bautista et al., 2007)(Colburn et al., 2007)(Dhaka et al., 2007) over a wide range of temperatures spanning

30 to 10°C. Activation of TRPM8 by icilin is reported to produce analgesia by activating central inhibitory pathways (Proudfoot et al., 2006).

The agonistic action of menthol has been extensively studied. TRPM8 permits the channeling of charged ions, usually calcium or potassium ions, to flow through cellular membranes to which it is attached when temperatures drop at or below 26 ± 2°C (Sarria & Gu, 2010). When temperatures fall below this region, the TRPM8 channel allows for membrane currents to increase at the peripheral nerve endings of cold-specific nonnociceptive afferents (A delta fibers) resulting in cold perception (Sarria & Gu, 2010)(Wasner, Schattschneider, Binder, & Baron, 2004). At those same temperatures, an associated intracellular increase in calcium ions is observed across the calcium permeable TRPM8 channel (Sarria & Gu, 2010). The literature shows that menthol acts within the presynaptic regions where TRPM8 channels are prevalent and the somatic sensory synapses connect primary afferent fibers and dorsal horn neurons in the spinal cord to the central nervous system (CNS).

In view of opposite actions exerted on TRPs that can have a beneficial effect, both on inflammation and inflammatory pain, addition of 3% menthol was thought to complement quite effectively Polyactil's effect on inflammation and pain, while making the application of cold inherent to every single treatment. Thus, menthol was added and Bioactil was created. A combined targeted effect on TRPs could be obtained by simply applying the new formulation to the affected area. It was conceived as a proof-of-concept formulation, which indeed it has proven exhaustively to be exquisitely effective for pain relief in wide human usage.

Further studies can be planned to demonstrate the molecular effects of the dual effect exerted by Bioactil in resolving inflammation and relieving inflammatory pain.

Since the inspiration for the formulation came from sports lesions, it was decided to name the new product Polyactil Sport. But because the formulation was not only extremely effective for sports injuries, but also for muscle and joint damaged from hard work or accidents, as well as being highly effective for conditions such as arthritis and fibromyalgia, it was decided to re-name the product Bioactil.

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