

Summary References Re: The Effects of NSAID on Tissue Healing

In the light of an increasing number of papers and publications that relate to the potentially inhibitory effect of non steroidal anti inflammatory drugs on musculoskeletal tissues healing and repair, a number of the key references and abstracts are presented below for consideration.

It is appreciated that this is not a comprehensive bibliography, but may assist those who wish to further evaluate the evidence in this critical area.

Allen, H. L., A. Wase, et al. (1980). "Indomethacin and aspirin: effect of nonsteroidal anti-inflammatory agents on the rate of fracture repair in the rat." *Acta Orthop Scand* 51(4): 595-600.

A total of 210 male Charles River CD rats, 45 days old, were subjected to a fracturing of the right radius and ulna by digital pressure while under ether anesthesia. These animals were then assigned randomly to dose groups (1, 2 or 4 mg/kg/day of indomethacin and 100, 200, or 300 mg/kg/day of aspirin) and were dosed for 21 days. Dose levels were chosen to provide approximately equipotent levels of the test compounds with the highest dose approaching toxicity. Radiographs were taken of control-rat fractures on days 8, 14, and 21. Three rats at 4 mg/kg/day of indomethacin died of interstitial perforation prior to scheduled sacrifice. On day 22, all remaining rats were sacrificed by exsanguination under anesthesia. Histologic sections of the radius and ulna were examined in random sequence without knowledge of the treatment regimen. A histologic grade based on the morphologic stage of fracture healing was given. There was a drug- and dose-related retardation of fracture healing, which was statistically significant at all dose levels of indomethacin and the highest level of aspirin, as compared to controls. Decreased mean grades in the groups given 100 and 200 mg/kg/day of aspirin, though not statistically significant, suggest a retarding effect on fracture healing at these levels also. No statistically significant changes in numbers of pseudoarthroses were found.

Almekinders, L. C. (1999). "Anti-inflammatory treatment of muscular injuries in sport. An update of recent studies." *Sports Med* 28(6): 383-8.

Stretch-induced muscle injuries or strains, muscle contusions and delayed-onset muscle soreness (DOMS) are common muscle problems in athletes. Anti-inflammatory treatment is often used for the pain and disability associated with these injuries. The most recent studies on nonsteroidal anti-inflammatory drugs (NSAIDs) in strains and contusions suggest that the use of NSAIDs can result in a modest inhibition of the initial inflammatory response and its symptoms. However, this may be associated with some small negative effects later in the healing phase. Corticosteroids have generally been shown to adversely affect the healing of these acute injuries. Animal studies have suggested that anabolic steroids may actually aid in the healing process, but clinical studies are not yet available and the exact role of these drugs has yet to be determined. Studies on anti-inflammatory treatment of DOMS have yielded conflicting results. However, the effect of NSAIDs on DOMS appears small at best. Future research may have to focus on different aspects of these injuries as the emphasis on anti-inflammatory treatment has yielded somewhat disappointing results.

Almekinders, L. C. and J. A. Gilbert (1986). "Healing of experimental muscle strains and the effects of nonsteroidal antiinflammatory medication." *Am J Sports Med* 14(4): 303-8.

The healing process of muscle strains and the effect of nonsteroidal antiinflammatory medication were studied using an experimental animal model. A standardized strain of the tibialis anterior muscle in adult male rats was produced by a controlled stretch of the muscle. Groups I and II underwent a unilateral strain of the tibialis anterior muscle and were immobilized in the postinjury period. The rats in Group II were fed piroxicam in the postinjury period. Group III underwent a sham procedure and were also immobilized. At 0, 2, 4, and 11 days postinjury both injured and contralateral control muscles were evaluated by determining tensile strength characteristics and histologic appearance. Group I showed a significant drop in maximum failure load to 25.7% of the control leg at Day 2 with a gradual return to the level of Group III at Days 4 and 11 (40.9% and 50.1%). Group II showed a similar drop but was still stronger than Group I at 2 days with 40.8% of the control leg and continued to drop until 4 days postinjury (33.7%). Histology showed a delay in inflammatory reaction and muscle regeneration in Group II. At 11 days both Groups I and II showed regenerated muscle fibers bridging the entire defect and an increase in endomyseal fibrosis. It is concluded that muscle strains continue to weaken in the early postinjury period. Non-steroidal antiinflammatory medication, such as piroxicam, might delay muscle regeneration.

Arceiz, E., A. Lanas, et al. (1997). "Platelet-derived growth factor reverses the effects induced by NSAIDs on ulcer healing." *Eur J Gastroenterol Hepatol* 9(12): 1179-84.

BACKGROUND: It is known that non-steroidal anti-inflammatory drug (NSAID) use delays the healing of peptic ulcers and that growth factors play an important role in the ulcer healing process. AIM: To evaluate the effect of platelet-derived growth factor (PDGF) in healing chronic gastric ulcers in rats treated with NSAIDs. METHODS: Chronic gastric ulcers were induced with acetic acid in male Wistar rats and then treated with either aspirin (100 mg/kg/day), indomethacin (2 mg/kg/day), PDGF-BB (0.1 nM/kg/day) or combinations. Gastric secretion and ulcer size, wound contraction, mucosal regeneration and cell proliferation were assessed in histological specimens. RESULTS: Both aspirin and indomethacin delayed the healing rate of gastric ulcers and reduced ulcer contraction, mucosal regeneration and cell proliferation. All these effects were completely reversed by oral treatment with PDGF-BB without affecting gastric acid secretion. CONCLUSION: Oral administration of PDGF accelerates ulcer healing and reverses the effects induced by NSAIDs on ulcer healing without affecting gastric secretion.

Bertsch, S. and F. Marks (1982). "A comparative study on wound-healing in neonatal and adult mouse epidermis in vivo." *Cell Tissue Kinet* 15(1): 81-7.

A cut made into the back skin of either newborn or adult mice evokes, at both ages, a hyperproliferative response in the epidermis. Differences in the reaction of neonatal as compared with adult epidermis are found in the spatial distribution of proliferative activity as well as in its time course. The response in adult mouse epidermis is inhibited by local application of indomethacin, whereas the response of the newborn epidermis is not.

Bo, J., E. Sudmann, et al. (1976). "Effect of indomethacin on fracture healing in rats." *Acta Orthop Scand* 47(6): 588-99.

The healing of closed, non-immobilized femoral fractures in rats was seriously impaired by indomethacin given orally at a dose of 2 mg/kg daily. The fracture haematomas were larger and disappeared later in the animals receiving indomethacin. Mechanical strength testing of fracture healing showed that maximal tensile strength, elastic stiffness and maximal bending moment between fragments were significantly diminished in the indomethacin-treated animals. Radiological examination showed a smaller amount of mineralized callus and a more pronounced angulation between the fragments in these animals than in the placebo-treated ones. Histological examination showed bridging between the fragments by callus tissue 24 days after fracture in placebo-treated animals, whereas indomethacin treatment was followed by histological findings resembling those seen in early pseudarthrosis development.

Bourne, M. S. (1980). "The effect on healing of analgesic and anti-inflammatory therapy." *Br J Sports Med* 14(1): 26.

Carlstedt, C. A. (1987). "Mechanical and chemical factors in tendon healing. Effects of indomethacin and surgery in the rabbit." *Acta Orthop Scand Suppl* 224: 1-75.

Carlstedt, C. A., K. Madsen, et al. (1986). "The influence of indomethacin on collagen synthesis during tendon healing in the rabbit." *Prostaglandins* 32(3): 353-8.

The influence of indomethacin on collagen synthesis in intact and healing plantaris longus tendons in the rabbit was investigated. Forty-four male New Zealand White rabbits were subjected to a standardized trauma (tenotomy + repair) on the left hindlimb. Half of the animals were subsequently treated with indomethacin, 10 mg/kg per day orally, and the other half with placebo. After 2 and 4 weeks the rabbits were injected intravenously with ³H-proline and killed 18 h later. Indomethacin affected the collagen metabolism differently depending on whether the tendons were involved in wound healing or not. In intact tendons the drug caused a small general inhibition of collagen synthesis. In the healing tendon there was a shift towards the synthesis of more insoluble collagen with little effect on the total synthesis. After 4 weeks there was also a slight but significant decrease in the amount of hydroxyproline in the most soluble collagen fraction from the tenotomized, indomethacin treated tendons.

Carlstedt, C. A., K. Madsen, et al. (1986). "The influence of indomethacin on tendon healing. A biomechanical and biochemical study." *Arch Orthop Trauma Surg* 105(6): 332-6.

The influence of indomethacin on the biomechanical and biochemical properties of tendons during their healing was investigated. In 68 New Zealand White rabbits a transverse tenotomy followed by repair with a criss-cross suture was performed in the plantaris longus tendon of the left hind limb. The

leg was immobilized for 4 weeks postoperatively in a long-leg plastic splint. Half of the animals were treated with indomethacin, 10 mg/kg/day orally, and the other half with placebo. After 4, 8, and 16 weeks of treatment the animals were killed and biomechanical and biochemical parameters were measured. After 16 weeks there was a significant increase in tensile strength in the indomethacin group. There were only small biochemical differences between the groups. However, there was a slight but significant decrease in the amount of soluble collagen in the indomethacin group. This may indicate a higher degree of cross-linkage following indomethacin treatment, which might explain the increased tensile strength.

Carlstedt, C. A., K. Madsen, et al. (1987). "The influence of indomethacin on biomechanical and biochemical properties of the plantaris longus tendon in the rabbit." *Arch Orthop Trauma Surg* 106(3): 157-60.

It has previously been reported that indomethacin inhibits fracture healing and heterotopic bone formation. Stimulated by these reports, we undertook the present investigation to study the influence of indomethacin on biomechanical and biochemical properties of the plantaris longus tendon in the rabbit. Sixty-eight New Zealand White rabbits were used for the experiment. Half of them were treated with indomethacin, 10 mg/kg orally a day, and the other half with placebo. After 4, 8, and 16 weeks of treatment biomechanical and biochemical variables were determined and compared between the two groups. After 16 weeks there was a significant increase in tensile strength in the group treated with indomethacin. There was no certain concomitant change in the total collagen content, the amounts of soluble and insoluble collagen, or the water content. Further investigations concerning the influence of indomethacin on tendon healing are indicated.

Dimar, J. R., W. A. Ante, et al. (1996). "The effects of nonsteroidal anti-inflammatory drugs on posterior spinal fusions in the rat." *Spine* 21(16): 1870-6.

STUDY DESIGN: This was a prospective study to determine the potential effects of indomethacin on spinal fusions in the rat. OBJECTIVES: To determine if indomethacin exerts a deleterious effect on spinal fusions in the rat model. SUMMARY OF BACKGROUND DATA: Nonsteroidal anti-inflammatory drugs are a class of compound that affect bone osteogenesis during fracture healing and heterotopic ossification. Spinal fusion is a process that occurs via osteogenesis and, therefore, may be similarly affected. METHODS: Thirty-nine adult, Sprague-Dawley rats underwent a three-level posterior spinal fusion. Fusion was performed using morselized autogenous vertebral bone graft obtained via caudectomy and stabilized using a cerclage wiring technique. The 39 rats were divided into two groups consisting of 17 study animals and 22 control animals. The control group was injected with 1.5 cc of 0.9 normal saline subcutaneously for 12 weeks, whereas the test animals were injected on an identical schedule using 3 mg/kg of indomethacin sodium salt. Two control animals died, and three animals in the treatment group died of drug-related complications. Twelve weeks after surgery, all animals were killed, and the involved spinal segments were evaluated by direct manual examination. A fusion was probable if the spinal segments exhibited decreased scaled micromotion. RESULTS: Sixty segmental levels in 20 control animals were assessed. Overall, 27 of 60 levels (45%) achieved fusion. In the indomethacin-treated group, 42 levels in 14 animals were evaluated. Overall, four of 42 levels (10%) achieved a fusion. Chi-square analysis demonstrated a significant difference ($P < 0.001$) between the control and indomethacin-treated groups. CONCLUSIONS: This study raises serious questions about the inhibitory effects of nonsteroidal anti-inflammatory drugs on spinal fusion. Clinically, the widespread use of nonsteroidal anti-inflammatory drugs in the postoperative period after spinal fusion may need to be avoided.

Diwan, P. V. and D. R. Kulkarni (1986). "Effects of nonsteroidal antiinflammatory agents (NSAIDs) on wound healing." *Indian J Exp Biol* 24(10): 640-3.

Dong, Y. L., R. Y. Fleming, et al. (1993). "Effect of ibuprofen on the inflammatory response to surgical wounds." *J Trauma* 35(3): 340-3.

Patients suffering severe trauma frequently become immunosuppressed following injury. This can predispose patients to infectious sequelae. Biochemically, these patients synthesize excessive quantities of cyclooxygenase products (prostaglandins). It has been hypothesized that the prostaglandins cause the immunosuppression and that inhibition of the cyclooxygenase enzyme could thus prevent the immunosuppression. We investigated the effect of the cyclooxygenase inhibitor ibuprofen on the inflammatory response. Rats were subjected to a 30% total body surface area burn and were administered either ibuprofen for a period of 7 days or 14 days, or were administered the carrier for 14 days. The rats were then killed and multiple immunologic variables were measured. Ibuprofen was found to decrease neutrophil chemiluminescence, lymphocyte blastogenesis, and helper/inducer T-lymphocyte infiltration of a sponge matrix model. The same ibuprofen protocol decreased survival in a

cecal ligation and puncture model. In conclusion, the cyclooxygenase enzyme system appears to produce metabolites essential for optimal survival following traumatic injury.

Durant, S., D. Duval, et al. (1989). "Effect of exogenous prostaglandins and nonsteroidal anti-inflammatory agents on prostaglandin secretion and proliferation of mouse embryo fibroblasts in culture." *Prostaglandins Leukot Essent Fatty Acids* 38(1): 1-8.

During wound healing, the positive and negative modulation of fibroblast proliferation may be due, in part, to the high prostaglandin concentration of the inflammatory exudates. In vitro, PGF2 alpha has been shown to stimulate, whereas PGE2 inhibits, the growth of different fibroblast cell lines. Therefore, we have investigated the effect of exogenous prostaglandins (PGs) and of various nonsteroidal anti-inflammatory drugs (NSAIDs) on the proliferation and the prostaglandin (PG) synthesis of normal mouse embryo fibroblasts. PGF2 alpha, 6-keto PGF1 alpha and PGE2 increase fibroblast proliferation. On the other hand, PGF2 alpha increases the synthesis of PGE2 and 6-keto PGF1 alpha while 6-keto PGF1 alpha solely inhibits PGF2 alpha release, PGE2 being inactive. The mouse embryo fibroblasts partially transform the prodrug sulindac sulfoxide in the sulfide form, which completely inhibits PG synthesis, as does indomethacin. In contrast, ibuprofen exerts a differential action, according to the type of PG measured. Among the NSAIDs tested, only sulindac (sulfoxide or sulfide) stimulates fibroblast proliferation and this effect appears independent of an alteration of PG synthesis. Therefore, in this model of normal mouse embryo fibroblasts, while endogenous prostaglandins are not involved in the control of cell proliferation, exogenous PGs have the ability to alter fibroblast growth and PG synthesis.

Dvivedi, S., S. M. Tiwari, et al. (1997). "Effect of ibuprofen and diclofenac sodium on experimental wound healing." *Indian J Exp Biol* 35(11): 1243-5.

Effects of frequently used anti-inflammatory drugs ibuprofen and diclofenac was studied on experimental wound healing in rats. These drugs impede tissue repair by virtue of retarding inflammation. There was 16-36% reduction in wound strength measured in terms of tensile strength in experimental rats. The detrimental effect of anti-inflammatory drugs was confirmed by histological examination of wound and by measuring dry granuloma weight.

Engesaeter, L. B., B. Sudmann, et al. (1992). "Fracture healing in rats inhibited by locally administered indomethacin." *Acta Orthop Scand* 63(3): 330-3.

We studied the inhibitory effect of indomethacin on fracture healing in 135 young, male rats after oral administration compared with local application into the fracture. A closed mid-diaphyseal fracture of the left femur was performed in all the rats. The fractures were not immobilized. In one experiment, half of the animals received indomethacin via a stomach tube (2 mg/kg/day) for 10 days; the controls received only the vehicle. In another experiment, 0.5 mg of indomethacin, contained in a bioerodible polyorthoester gel, was injected into the fracture area in half the rats; in the controls, only the gel was injected. In both experiments, random animals were killed on Days 0, 5, 10, and 20. As assessed by radiographs and manual testing, the same inhibition of fracture healing was found regardless of whether indomethacin was given orally or locally. However, the amount of indomethacin that was applied locally was only one fourth of the total dose given orally; no indomethacin was detected in the serum.

Fang, C., J. W. Alexander, et al. (1983). "Failure of topical prostaglandin inhibitors to improve wound healing following deep partial-thickness burns." *J Trauma* 23(4): 300-4.

The present study was designed to determine the efficacy of topical inhibitors of prostaglandins on wound healing. Two uniform deep partial-thickness burns were inflicted on mirror-image areas of guinea pig backs by an aluminum template heated to 75 degrees C and applied for 10 seconds. Indomethacin was tested extensively in a wide range of concentrations in groups of six or more animals each. The healing rates measured at 21 days postburn showed that topical indomethacin at each concentration tested was not effective for improving wound healing. In fact, the treated sites were consistently worse than the control sites. Moreover, the drug adversely affected the healing process proportional to the concentration and was associated with death, which was related to perforations of the GI tract. Also, the India ink filling in the dermal microcirculation was no better in the experimental wounds than in the controls. The evaluations for hair growth were definitely in favor of the controls. The other tested inhibitors, ibuprofen, flurbiprofen, tolmetin, zomepirac, piroxicam, and dipyridamole, also failed to show any benefit.

Giannoudis, P. V., D. A. MacDonald, et al. (2000). "Nonunion of the femoral diaphysis. The influence of reaming and non-steroidal anti-inflammatory drugs." *J Bone Joint Surg Br* 82(5): 655-8.

We assessed factors which may affect union in 32 patients with nonunion of a fracture of the diaphysis of the femur and 67 comparable patients whose fracture had united. These included gender, age, smoking habit, the use of non-steroidal anti-inflammatory drugs (NSAIDs) the type of fracture (AO classification), soft-tissue injury (open or closed), the type of nail, the mode of locking, reaming non-reaming, infection, failure of the implant, distraction at the fracture site, and the time to full weight-bearing. Patients with severe head injuries were excluded. Both groups were comparable with regard to gender, Injury Severity Score and soft-tissue injury. There was no relationship between the rate of union and the type of implant, mode of locking, reaming, distraction or smoking. There were fewer cases of nonunion in more comminuted fractures (type C) and in patients who were able to bear weight early. There was a marked association between nonunion and the use of NSAIDs after injury ($p = 0.000001$) and delayed healing was noted in patients who took NSAIDs and whose fractures had united.

Hogevold, H. E., B. Groggaard, et al. (1992). "Effects of short-term treatment with corticosteroids and indomethacin on bone healing. A mechanical study of osteotomies in rats." *Acta Orthop Scand* 63(6): 607-11.

We studied the effects of short-term therapy with methylprednisolone and indomethacin on healing of intramedullary pinned osteotomies of the femur in rats. When the osteotomy was complete and healing occurred under unstable conditions with callus formation, indomethacin inhibited healing when estimated by mechanical tests of bending moment, energy expenditure before refracture, and bending rigidity 6 weeks after surgery. No inhibitory effects were seen following corticosteroid treatment. When the osteotomy was incomplete and healing occurred under stable conditions, similar tendencies were observed. Thus, short-term medication with indomethacin inhibits fracture healing. This was not the case with short-term methylprednisolone.

Jones, M. K., H. Wang, et al. (1999). "Inhibition of angiogenesis by nonsteroidal anti-inflammatory drugs: insight into mechanisms and implications for cancer growth and ulcer healing." *Nat Med* 5(12): 1418-23.

Angiogenesis, the formation of new capillary blood vessels, is essential not only for the growth and metastasis of solid tumors, but also for wound and ulcer healing, because without the restoration of blood flow, oxygen and nutrients cannot be delivered to the healing site. Nonsteroidal anti-inflammatory drugs (NSAIDs) such as aspirin, indomethacin and ibuprofen are the most widely used drugs for pain, arthritis, cardiovascular diseases and, more recently, the prevention of colon cancer and Alzheimer disease. However, NSAIDs produce gastroduodenal ulcers in about 25% of users (often with bleeding and/or perforations) and delay ulcer healing, presumably by blocking prostaglandin synthesis from cyclooxygenase (COX)-1 and COX-2 (ref. 10). The hypothesis that the gastrointestinal side effects of NSAIDs result from inhibition of COX-1, but not COX-2 (ref. 11), prompted the development of NSAIDs that selectively inhibit only COX-2 (such as celecoxib and rofecoxib). Our study demonstrates that both selective and nonselective NSAIDs inhibit angiogenesis through direct effects on endothelial cells. We also show that this action involves inhibition of mitogen-activated protein (MAP) kinase (ERK2) activity, interference with ERK nuclear translocation, is independent of protein kinase C and has prostaglandin-dependent and prostaglandin-independent components. Finally, we show that both COX-1 and COX-2 are important for the regulation of angiogenesis. These findings challenge the premise that selective COX-2 inhibitors will not affect the gastrointestinal tract and ulcer/wound healing.

Jones, P. G. (1999). "Analgesia in soft-tissue injury: current practice in Auckland is not supported by the available evidence." *N Z Med J* 112(1097): 376-9.

AIMS: To document current prescribing habits and attitudes of doctors in the Auckland region towards analgesic medication for soft-tissue injury and determine whether the available evidence supports this practice. METHOD: A survey of 573 doctors in the Auckland region was conducted. There was a 71.4% response rate. The clinical and experimental evidence concerning non-steroidal, anti-inflammatory (NSAID) use in soft-tissue injury was reviewed. The side-effect profiles of NSAIDs were reviewed, with emphasis on the incidence of gastrointestinal side-effects when NSAIDs are prescribed for short periods and evidence implicating adverse renal effects on healthy exercising adults. RESULTS: Most doctors ranked NSAIDs more effective than paracetamol (70.4%, $p < 0.01$). NSAIDs were the most prescribed single analgesic agents (47.8%, $p < 0.0001$). Diclofenac was the NSAID of choice for 69.8% of doctors, who used NSAIDs ($p < 0.001$). The incidence of gastrointestinal side-effects for short-term use of NSAIDs in acute soft tissue was 11%. CONCLUSION: The available evidence does not support the belief by the doctors surveyed that NSAIDs are more effective than paracetamol in soft-tissue injury. NSAIDs delay, but do not prevent the inflammatory response in

injured tissue and may expose athletes to an increased risk of re-injury by delaying healing. Significant adverse effects do occur in previously healthy patients who receive NSAIDs.

Keller, J. (1996). "Effects of indomethacin and local prostaglandin E2 on fracture healing in rabbits." *Dan Med Bull* 43(4): 317-29.

Keller, J., C. Bungler, et al. (1987). "Bone repair inhibited by indomethacin. Effects on bone metabolism and strength of rabbit osteotomies." *Acta Orthop Scand* 58(4): 379-83.

We measured mineral content, maximum bending strength, and regional blood flow after tibial osteotomy fixed with a small metal plate in 38 rabbits. Half of the animals were treated with indomethacin (10 mg/kg/day) while the other half served as controls. After 2 and 6 weeks, the bone mineral content and maximum bending strength were lower in the indomethacin group when compared with the controls. Compared with the controls, the blood flow at the osteotomy site was decreased after 2 weeks and increased after 6 weeks in the indomethacin-treated animals. Inhibition of blood flow increase by indomethacin medication in the early period following osteotomy, as well as retarded bone healing, are probably caused by inhibition of the inflammatory reaction.

Keller, J., P. Kjaersgaard-Andersen, et al. (1990). "Indomethacin and bone trauma. Effects on remodeling of rabbit bone." *Acta Orthop Scand* 61(1): 66-9.

The influence of indomethacin on remodeling activity in normal trabecular and cortical bone and its influence on cortical bone close to a midtibial drill hole, 2 mm in diameter were histomorphometrically evaluated. Eight rabbits were treated with indomethacin (12.5 mg/kg/day), and another 8 rabbits served as controls. After 3 days, the mean plasma indomethacin level was 542 ng/mL, resulting in an almost complete inhibition of prostaglandin synthesis as reflected by the serum levels. In the control rabbits the remodeling activity after 6 weeks was increased 1 mm away from the drill hole but not at 3 and 8 mm. In conclusion, indomethacin had no effect on the activated remodeling process in cortical bone neighboring a small drill hole or on remodeling in nontraumatized cortical and cancellous bone. This suggests that the inhibitory effect of indomethacin on the remodeling process following local trauma to bone depends on the extent of the trauma.

Khan, M. I. (1997). "Fracture healing: role of NSAIDs." *Am J Orthop* 26(6): 413.

Kumar, A., M. Rao, et al. (1988). "Zinc incorporation reverses suppressant effect of ibuprofen on wound healing." *Indian J Exp Biol* 26(6): 483-5.

Lane, N., T. Coble, et al. (1990). "Effect of naproxen on cancellous bone in ovariectomized rats." *J Bone Miner Res* 5(10): 1029-35.

Nonsteroidal anti-inflammatory drugs (NSAIDs) affect bone metabolism in vitro and in vivo. They delay but do not alter the outcome of healing processes in bone. In some bone loss models, they block bone resorption and slow the rate of loss. We studied the effect of naproxen, a potent NSAID, on cancellous bone of the proximal tibial metaphysis of 6-month-old adult female ovariectomized rats. Animals were ovariectomized, divided into groups, and fed standard diets differing only in naproxen content for 42 days. The rats of the groups ate 2.0, 5.5, 12.7, and 32 mg naproxen per kg body weight per day, respectively. Serum levels of naproxen were determined. Bone volume, mineralizing surface, osteoblast activity, osteoclast surface, and bone resorption rate were determined by bone histomorphometric techniques. The rats' dose-related serum naproxen levels ranged from 4 to 28 micrograms/ml. Naproxen inhibited up to 70% of the bone loss occurring after ovariectomy at a serum level of 4 micrograms/ml. We deduced that naproxen blocked bone resorption in ovariectomized rats by slowing osteoclast activity at all doses. In contrast, naproxen slowed bone formation only at serum levels greater than 20 micrograms/ml in ovariectomized rats. These findings may have clinical relevance in helping to prevent postmenopausal bone loss in women.

Moorman, C. T., U. Kukreti, et al. (1999). "The early effect of ibuprofen on the mechanical properties of healing medial collateral ligament." *Am J Sports Med* 27(6): 738-41.

We tested the hypothesis that injured ligaments in rabbits treated with ibuprofen would have decreased values of mechanical properties compared with the values of those treated with a placebo. In 24 New Zealand White rabbits, the medial collateral ligament of one hindlimb was ruptured; the contralateral ligament served as an internal control. The rabbits were treated orally, twice daily, with a 14-day course of either 35 mg of ibuprofen per kilogram of body weight or a placebo. The rabbits were sacrificed at 14 or 28 days, and the ligaments were tested in tension to failure at 0.15 mm/sec. There was no statistically significant difference in the values of mechanical properties of ligaments from rabbits treated with ibuprofen versus those treated with placebo at either 14 or 28 days after injury. Our

findings suggest that there is no early deleterious effect of a short course of ibuprofen on the mechanical behavior of medial collateral ligaments.

Muscara, M. N., W. McKnight, et al. (2000). "Wound collagen deposition in rats: effects of an NO-NSAID and a selective COX-2 inhibitor." *Br J Pharmacol* 129(4): 681-6.

Selective cyclo-oxygenase (COX)-2 inhibitors and nitric oxide-releasing nonsteroidal anti-inflammatory drugs (NSAIDs) exhibit reduced toxicity in the gastrointestinal tract, but may affect wound healing in other tissues. In this study, we have compared the effects of a selective COX-2 inhibitor (celecoxib), a nitric-oxide releasing derivative of naproxen (HCT-3012) and naproxen in a model of wound collagen deposition in the rat. Polyvinyl alcohol sponges were implanted subcutaneously in rats. The rats were treated daily for 5 days with the test drugs at equieffective anti-inflammatory doses. Naproxen (10 mg kg⁻¹) significantly decreased (45%) collagen deposition at the wound site relative to the vehicle-treated control group. In contrast, HCT-3012 (14.5 mg kg⁻¹) significantly increased (62%) collagen deposition, while celecoxib (10 mg kg⁻¹) had no effect. Naproxen and HCT-3012 suppressed prostaglandin (PG) E₂ levels at the wound site and whole blood thromboxane synthesis to similar degrees. Celecoxib had no significant effect on wound fluid PGE₂ levels, but slightly reduced whole blood thromboxane synthesis (by 17%). COX-1 mRNA and protein were expressed in the wound exudate, the skin surrounding the wound and in normal skin. In contrast, COX-2 mRNA, but not protein, was expressed in wound and normal skin. These results demonstrate that HCT-3012 can significantly enhance collagen deposition at a wound site, despite inhibiting prostaglandin synthesis to the same extent as the parent drug. Nitric oxide-releasing NSAIDs may represent a safer alternative to standard NSAIDs for use as anti-inflammatory and analgesic agents by post-surgery patients.

Obeid, G., X. Zhang, et al. (1992). "Effect of ibuprofen on the healing and remodeling of bone and articular cartilage in the rabbit temporomandibular joint." *J Oral Maxillofac Surg* 50(8): 843-9; discussion 849-50.

The purpose of this study was to determine the effect of ibuprofen on the healing and remodeling of bone and cartilage in the temporomandibular joint of the rabbit. Forty-two rabbits were operated on to create a groove and a hole in the articular surface of both the right and left mandibular condyles. Following surgery, the animals were divided into three groups. Group A (12 rabbits) was used as a control and the animals did not receive any medication. Group B (15 rabbits) was given a daily dose of 17 mg/kg of ibuprofen. Group C (15 rabbits) was given a daily dose of 34 mg/kg of ibuprofen. All animals were killed after 4 weeks. The 84 condyles were examined clinically and histologically. Statistical analysis showed a highly significant difference in the healing of bone and cartilage between groups A and C (P less than .01) and a significant difference between groups A and B (P less than .05). The results of this study indicate that ibuprofen has an adverse effect on the healing of bone and cartilage in the temporomandibular joint of the rabbit.

Reynolds, J. F., T. D. Noakes, et al. (1995). "Non-steroidal anti-inflammatory drugs fail to enhance healing of acute hamstring injuries treated with physiotherapy." *S Afr Med J* 85(6): 517-22.

The effects of two non-steroidal anti-inflammatory drugs (NSAIDs), meclofenamate and diclofenac, in combination with physiotherapy modalities on the rate of healing of acute hamstring muscle tears were studied in a double-blind, placebo-controlled trial. Forty-four of the 75 patients with this injury recruited were assessed and randomly allocated to one of three treatment groups: meclofenamate (100 mg 3 times a day), diclofenac (50 mg 3 times a day) and placebo. All patients received the same intensive physiotherapy treatment over the 7-day treatment period. Patient assessments were performed on days 1, 3 and 7 of the 7-day study period and included pain assessment (visual analogue scale), swelling measurement (thigh circumference measurement at the site of the muscle tear) and isokinetic muscle performance testing. Treatment produced a significant improvement in all measurements in all groups, but there was no difference in any measurement between groups. However, when only the more severe injuries were analysed, the reported pain score at day 7 was significantly lower in the placebo group than in either the meclofenamate group or the diclofenac group (P < 0.05). Hence this study did not find any additive effect on the healing of acute muscle injuries when meclofenamate or diclofenac was added to standard physiotherapeutic modalities. The study therefore does not support the use of NSAIDs in the treatment of acute hamstring muscle injuries.

Tornkvist, H. and T. S. Lindholm (1980). "Effect of ibuprofen on mass and composition of fracture callus and bone. An experimental study on adult rat." *Scand J Rheumatol* 9(3): 167-71.

Sixty adult male rats were used to study the effect of the anti-inflammatory drug Ibuprofen on fracture callus and perpendicular skeleton. After experimental periods of 3 and 9 weeks, fracture callus and

both fractured and unfractured tibiae were examined with respect to bone mass and composition and ^{45}Ca metabolism. No significant changes were found in the composition of fracture callus during treatment. Significantly diminished parameters of both fractured and unfractured tibia were observed for wet and dry weights, ash content, and organic matter after 3 weeks but the bone mass had become almost restored and the changes were non-significant during treatment 9 weeks following fracturing. The ^{45}Ca activity was elevated significantly in fracture callus and fractured tibia 3 weeks after fracturing but had definitely declined to physiological levels at 9 weeks. Serum ^{45}Ca activity was significantly elevated during Ibuprofen treatment. The findings support the concept that Ibuprofen lessens the bone mass and composition of both fractured and unfractured tibia and also activates the calcium metabolism in fracture callus. In the long run, however, this effect is weakened and the bone changes are become almost normal. Some explanations regarding these short-term effects of Ibuprofen are discussed.