Review articles

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THE FIGHT AGAINST RHEUMATISM: FROM WILLOW BARK TO COX-1 SPARING DRUGS

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Man has been fighting rheumatism for thousands of years. Early therapy began with the use around the world of decoctions or extracts of herbs or plants such as willow bark or leaves. Most or all of these turned out to contain salicylates. The first record was about 3,500 years ago in the Ebers papyrus. Hippocrates, Celsus, Pliny the Elder, Dioscorides and Galen all recommended decoctions containing salicylate for rheumatic pain. A country parson, the Reverend Edward Stone of Chipping Norton in Oxfordshire, made the first “clinical trial” of willow bark (1). He was surprised by its bitter taste, which reminded him of cinchona bark (containing quinine), then being used to treat malaria. He harvested a pound of willow bark, dried it, pulverized it and dispersed it in tea, small beer or water. He found in 50 patients that doses of 1 dram (1.8g) cured their fever. He concluded “I have no other motives for publishing this valuable specific, than that it may have a fair and full trial in all its variety of circumstances and situations, and that the world may reap the benefits accruing from it”. Salicylic acid was chemically synthesised in 1860 by Kolbe in Germany and its ready supply led to even more extended usage as an external antiseptic, as an antipyretic and in the treatment of rheumatism.

Key words: rheumatism, aspirin, prostaglandins, COX-1, COX-2, NSAIDS, selective COX-2 inhibitor

INTRODUCTION

Aspirin was born just over 100 years ago. Felix Hoffman was a young chemist working for Bayer in Germany. Legend has it that his father was taking salicylate for his severe rheumatism and urged Felix to find a more palatable form. Nowadays, it is easy to disguise the taste of a medicine by putting a hard sugar coating on the tablet, but neither tablets nor coatings were available. Instead, Felix made acetylsalicylic acid, named as aspirin. Bayer’s Research Director, Dr Heinrich Dreser, tested aspirin in animals and found it to be antipyretic, analgesic and anti-inflammatory. He recognized that he had an important new drug on his hands and introduced it to the market as
a powder in 1899 and as tablets soon thereafter. The wishes of the Reverend Edward Stone have certainly been realised; world production of acetylsalicylic acid is now estimated at 50 thousand tons a year, with an average consumption of about 80 tablets per person per year. Without the discovery in recent years of many other aspirin-like drugs, such as ibuprofen, the fenamates, indomethacin and naproxen, consumption would have surely been very much higher.

Despite the diversity of their chemical structures, these non-steroid anti-inflammatory drugs (NSAIDS) all share the same therapeutic properties, alleviating the swelling, redness and pain of inflammation, reducing fever and curing headaches. Importantly, they also share to a greater or lesser extent the same group of side effects, including interfering with the birth process and damaging the kidney. However, the most troublesome side effect is on the stomach. Indeed, epidemiological studies have characterised the degree of gastric damage caused by different compounds (2). An estimated 34—-46% of patients on NSAID therapy will have some form of gastrointestinal adverse events (3). In the USA alone, some 100,000 patients on NSAIDs are hospitalised each year because of perforations, ulcers or bleeding in the stomach (PUBs) (4) and about 15,000 of these die in intensive care. Of course, these hospitalisations only represent the extreme of gastric irritation, which ranges from mild dyspepsia all the way through to PUBs. Even ibuprofen, recognised as one of the mildest gastric irritants, causes problems in a significant proportion of patients. Clearly, there is dramatic need for anti-inflammatory drugs that do not affect the stomach.

**Discovery of the mode of action of aspirin**

For many years pharmacologists and biochemists searched for a common mode of action without finding a generally acceptable scientific explanation. In the late 1960s I was working on a newly discovered group of chemical mediators called the prostaglandins. Many types of chemical or mechanical stimuli cause their synthesis and release in different parts of the body. Interestingly, one or more of this group of lipid-derived mediators caused pain, swelling and redness. They also contracted many kinds of smooth muscle, including that of the uterus. In addition, they increased renal blood flow and reduced gastric acid secretion. These were all activities with which aspirin interfered in some way. Could it be that aspirin was blocking the biosynthesis of prostaglandins?

I tested this idea immediately in vitro, using as a source of prostaglandin synthase the supernatant of a broken cell homogenate from guinea pig lung (5). There was a dose-dependent inhibition of prostaglandin formation by aspirin, salicylate and indomethacin but not by morphine (6). Two other reports (7, 8) from my laboratory in the same issue of Nature lent support to these findings. The discovery that each and every chemically diverse member of this large
group of drugs all act by inhibiting the key enzyme in prostaglandin biosynthesis (which we now call cyclooxygenase or COX) provided a unifying explanation for their therapeutic actions and shared side effects (for reviews see refs 9—11). This theory became well accepted, although there was always the puzzle as to why the different drugs, at therapeutic concentrations, varied widely in the severity of their side effects.

Some companies found new compounds that were anti-inflammatory, but were developed because they were less damaging to the stomach, usually tested in rats. Meloxicam, nimesulide and etodolac, all now recognised as selective COX-2 inhibitors, were discovered in this way.

The Discovery of COX-2

There were various clues in the literature suggesting that there may be a second COX enzyme. As early as 1972, Smith and Lands (12) and Flower and Vanc (13) speculated on the existence of isoenzymes. Maddox (14) showed the presence of two separate prostaglandin synthase complexes in sheep vesicular tissue. Others (15—17) also suggested two distinct forms of COX. Rosen et al. (1989) (18) were studying the regulation of cyclooxygenase in cultures of epithelial cells from trachea and found an increase in activity of COX during prolonged cell culture. The increase in activity was not accounted for by the increase in 70 kDa cyclooxygenase protein, nor by the mRNA of 2.8 kb. They did find a second mRNA of 4.0 kb and suggested that their evidence was consistent with the 4.0 kb mRNA being derived from a distinct cyclooxygenase-related gene which encode for a protein with COX activity.

Needleman and his group (19—21) reported that bacterial lipopolysaccharide (LPS) increased the synthesis of prostaglandins in human monocytes in vitro and in mouse peritoneal macrophages in vivo. This increase, but not the basal level of enzyme, was inhibited by dexamethasone and associated with de novo synthesis of new COX protein. This further gave rise to the concept of “constitutive” and “inducible” forms of COX.

The breakthrough came from molecular biologists outside the field of prostaglandins. Simmons and his colleagues in 1991 (22, 23) were studying early response genes and discovered an inducible second form of COX in chicken embryo cells. It was encoded by a 4.1 kb mRNA similar in size to that reported by Rosen et al. (18). They cloned the gene, deduced the protein structure and found it homologous to COX, but to no other known protein.

This work was closely followed by Herschmann and his colleagues (24), who found a similar gene in the mouse, as did Simmons et al. (25), O’Banion et al. (26) and Siros and Richards (27). Both enzymes have a molecular weight of 71 Kd and the amino acid sequence of the cDNA for COX-2 shows a 60% homology with the sequence of the non-inducible enzyme. The mRNA for the
inducible enzyme approximates 4.5 kb and that of the constitutive enzyme, 2.8 kb. The inhibition by glucocorticoids of the expression of COX-2 is an additional aspect of the anti-inflammatory action of the corticosteroids. The levels of COX-2, normally very low in cells, are tightly controlled by a number of factors including cytokines, intracellular messengers and by the availability of substrate.

Thus, the constitutive isoform COX-1 has clear physiological functions. Its activation leads, for instance, to the production of prostacyclin which when released by the endothelium is anti-thrombogenic (28) and when released by the gastric mucosa is cytoprotective (29). The second isoform, COX-2, is inducible in a number of cells by pro-inflammatory stimuli (30). Since COX-2 is induced by inflammatory stimuli and by cytokines in migratory and other cells it was attractive to suggest, as I and others did in 1993 (31, 32), that the anti-inflammatory actions of NSAIDs are due to the inhibition of COX-2, whereas the unwanted side effects such as irritation of the stomach mucosa and toxic effects on the kidney are due to inhibition of the constitutive enzyme, COX-1. This very important hypothesis is now well supported, not only by a wealth of data on COX-2 inhibitors in animal tests, but also by the better tolerability in man of selective COX-2 inhibitors (see below).

The design of selective COX-2 inhibitors

Two brilliant scientists led the field, racing with their teams to produce selective COX-2 inhibitors. Tony Ford-Hutchinson, then at Merck Frosst in Canada and Phil Needleman, then Research Director at Monsanto Searle (now Pharmacia) committed substantial resources to the projects. The results were the marketing within ten years of rofecoxib (VIOXX) by Merck and of celecoxib (Celebrex) by Searle. Favourable clinical trial results are already available for meloxicam (Mobic) (33—35): both rofecoxib and celecoxib have shown similar superiority to the NSAIDs from the safety point of view in extensive clinical trials yet to be published. Rofecoxib and celecoxib are effective analgesics in man for moderate to severe pain following tooth extraction (36, 37).

Measuring the effects of aspirin-like drugs on the two enzymes

Mitchell et al. (31) were the first to measure the effects of NSAIDs on both COX-1 and COX-2. Recently, several methods have been published to determine the relative actions of NSAIDs on COX-1 and COX-2. These range from isolated enzymes (now usually recombinant human enzymes) through whole cell preparations in vitro to the human whole blood assay (see Ref 38 for discussion of relative merits). The isolated enzyme assays give the highest ratios for COX-2/COX-1. For instance, meloxicam has a ratio of 100, whereas celecoxib and rofecoxib have ratios of more than 1000 in favour of COX-2 (39).
However, such assays do not take into account the avid, but variable binding of some of these drugs to plasma protein and other aspects of the kinetics of drug distribution. It is now generally accepted that the human whole blood assay, first described by Patrono and colleagues (40) best reflects activity in vivo in man. The activity of compounds on COX-2 is measured in the platelets of the blood sample and that on COX-2 in white cells induced over 24 hours to express COX-2 by LPS or a cytokine. A useful modification by Warner et al. (41) reduces the time to measure COX-2 effects down to 1.5 hours.

The advantages of these methods are that they use human cells that importantly are in a physiological environment (plasma), which automatically takes any protein binding into account. What is more, the assays give reproducible results between various laboratories. For significant differences between drugs, changes in ratio of an order of magnitude are needed. Interestingly, the ratios change substantially in this assay. In our hands (41), for example, meloxicam has a ratio in favour of COX-2 of about 5, celecoxib is hardly different at 10, whereas rofecoxib, has a ratio of >60 (see Table 1).

Table 1. The COX-2/COX-1 ratios for some NSAIDs and selective COX-2 inhibitors in whole blood assays in different laboratories. The data from Warner et al. (41) is for the modified whole blood assay (a) and for the usual whole blood assay (b).

<table>
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<tr>
<th>DRUG</th>
<th>Warner et al. (41) (a)</th>
<th>Warner et al. (41) (b)</th>
<th>Patrignani et al. (40)</th>
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Selective COX-2 inhibitors in current therapeutic use

Meloxicam, nimesulide and etodolac were identified in the 1980s as potent anti-inflammatory drugs with low ulcerogenic activity in the rat stomach. In some instances, this was also shown to parallel low activity against prostaglandin synthesis in the rat stomach. After the characterisation of the COX-2 gene, these three drugs were each found selectively to inhibit COX-2 rather than COX-1 (see Table 1).

Meloxicam, which has a selectivity towards COX-2 of about 5 in the human whole blood assay is marketed around the world for use in rheumatoid arthritis and osteoarthritis. In double blind trials (33—35) in many thousands of patients with osteoarthritis, meloxicam in doses of 7.5 mg or 15 mg once daily compared in efficacy with standard NSAIDs such as naproxen 750—1000 mg, piroxicam 20 mg or diclofenac 100 mg. Both doses of meloxicam produced significantly fewer gastrointestinal adverse effects than the standard NSAIDs (p < 0.05). Discontinuation of treatment due to gastrointestinal side effects was also significantly less frequent with meloxicam. Perforations, ulcers and bleedings occurred in fewer meloxicam-treated patients than in patients treated with piroxicam, diclofenac or naproxen. The frequency of adverse events with meloxicam was significantly less at p < 0.05 when compared to piroxicam and naproxen. These large-scale clinical trials with a selective COX-2 inhibitor add weight to the concept that the sparing of COX-1 inhibition will reduce gastric damage.

Etodolac is marketed in Europe and North America for the treatment of osteoarthritis and rheumatoid arthritis. It has about five fold selectivity for COX-2 in human whole blood (42). In healthy human volunteers, etodolac twice daily did not suppress gastric mucosal prostaglandin production and caused less gastric damage than naproxen (43). Patients with osteoarthritis or rheumatoid arthritis obtained relief from symptoms equal to other commonly used NSAIDs with etodolac, but with a lower incidence of serious gastrointestinal toxicity (44).

Nimesulide is currently sold in Europe and South America for the relief of pain associated with inflammatory conditions. It is a selective inhibitor of COX-2 with about five fold greater potency against this enzyme than against COX-1 in the human whole blood assay (Table 1). In limited clinical trials for its use in acute and chronic inflammation in patients it was more effective than placebo or had comparable anti-inflammatory activity to established NSAIDs. Interestingly, nimesulide seems safe to use in aspirin-sensitive asthmatics. Several recent studies in NSAID-intolerant asthmatic patients demonstrated that therapeutic doses of nimesulide did not induce asthmatic attacks while high doses of 400 mg only precipitated mild asthma in 10% of patients (45). Perhaps aspirin-induced asthma is associated with COX-1 inhibition?
Interestingly, COX-2 is the constitutive and dominant form of the enzyme in human cultured lung epithelial cells (46).

A recent epidemiological study (47) identified 1505 patients with upper gastrointestinal tract bleeding. It showed nimesulide to have a similar relative risk to that of naproxen (4.4 times control) and more than diclofenac (2.7 times control). Clearly, other factors are also involved, such as frequency of dosage etc. As with other NSAIDs, nimesulide is used in different dosages and when these are separated, the higher doses give a much higher relative risk.

Constitutive COX-2

Maintenance of kidney function both in animal models of disease states and in patients with congestive heart failure, liver cirrhosis or renal insufficiency, is dependent on vasodilator prostaglandins. These patients are, therefore, at risk of renal ischaemia when prostaglandin synthesis is reduced by NSAIDs. Synthesis of PGE$_2$ is mainly by COX-1, although there are discrete cells in the macula densa that contain constitutive COX-2 (48, 49). Prostacyclin, made by constitutive COX-2 may drive the renin-angiotensin system (49) Schneider and Stahl (50) have reviewed this rapidly evolving field.

Fitzgerald's group (51) compared the renal effects of the non-selective COX inhibitor, indomethacin with those of the COX-2 inhibitor, rofecoxib and with placebo in healthy older adults over two weeks treatment. Both active regimes were associated with a transient but significant decline in urinary sodium excretion during the first 72 hours. The glomerular filtration rate (GFR) was decreased by indomethacin but not changed significantly by rofecoxib. Thus, acute sodium retention by NSAIDs in healthy adults is mediated by inhibition of COX-2, whereas depression of GFR is due to inhibition of COX-1.

The urinary excretion of the prostaglandin metabolite 2,3-dinor-6keto prostaglandin F$_{1\alpha}$ was decreased by both rofecoxib and indomethacin, but not by placebo. The implication of this is that the endothelial cell uses COX-2 to make prostacyclin, this enzyme is possibly induced by the shear stress in the arterial wall, rather than being present constitutively.

COX-1 is found in neurones throughout the brain but it is most abundant in forebrain where prostaglandins may be involved in complex integrative functions such as control of the autonomic nervous system and in sensory processing. COX-2 mRNA is induced in brain tissue and in cultured glial cells by pyrogenic substances such as LPS, IL-1 or TNF (52). However, low levels of COX-2 protein and COX-2 mRNA have been detected in neurones of the forebrain without previous stimulation by pro-inflammatory stimuli. These „basal” levels of COX-2 are particularly high in neonates and are probably induced by physiological nervous activity. Intense nerve stimulation, leading to seizures, induces COX-2 mRNA in discrete neurones of the hippocampus (53).
whereas acute stress raises levels in the cerebral cortex. COX-2 mRNA is also constitutively expressed in the spinal cord of normal rats and may be involved with processing of nociceptive stimuli (54). Endogenous, fever-producing PGE$_2$ is thought to originate from COX-2 induced by LPS or IL-1 in endothelial cells lining the blood vessels of the hypothalamus (52).

Li et al. (55) tested the effects of LPS in producing a fever in knockout mice. Wild type, and COX-1$_{(+/-)}$ and COX-1$_{(-/-)}$ mice all responded to LPS with a 1°C rise in core temperature within 1 hour: the fever gradually abated over the next 4 hours. By contrast, COX-2$_{(+/-)}$ and COX-2$_{(-/-)}$, mice displayed no temperature rise after LPS. Thus, COX-2 is necessary for the production of fever produced by LPS. A corollary of this finding is that there is unlikely to be a COX-3 through which paracetamol brings down a fever. Furthermore, Oates et al. (56) has recently shown that in HUVEC cells in culture, paracetamol inhibits COX-2 with an IC$_{50}$ of 66um, well within the therapeutic range in humans. Furthermore, the selective COX-2 inhibitor, rofecoxib, is a potent antipyretic agent in man (57).

**Nomenclature**

Merck and Searle refer to their new COX-2 inhibitors as “specific”, arguing that at therapeutic doses, there is only inhibition of COX-2 and not of COX-1. Pharmacologists use the word “specific” far more rigorously, and „selective” is a more appropriate description. Even more accurate would be „COX-1 sparing drugs”.

**FUTURE THERAPEUTIC USES FOR SELECTIVE COX-2 INHIBITORS**

**Premature labour**

Prostaglandins (PGF$_{2a}$) induce uterine contractions during labour. NSAIDs such as indomethacin will delay premature labour by inhibiting the production of prostaglandins, but will at the same time cause early closure of the ductus arteriosus and reduce urine production by the fetal kidneys (58). The delay in the birth process is most likely due to inhibition of COX-2 since mRNA for COX-2 increases substantially in the amnion and placenta immediately before and after the start of labour (59), whereas the side effects on the fetus are due to inhibition of COX-1. One cause of pre-term labour could be an intra-uterine infection resulting in release of endogenous factors that increase prostaglandin production by up-regulating COX-2 (ref 60). Nimesulide reduces prostaglandin synthesis in isolated fetal membranes and has been used successfully for a prolonged period to delay premature labour without manifesting the side effects of indomethacin on the fetus (58).
Colon cancer

Epidemiological studies have established a strong link between ingestion of aspirin and a reduced risk of developing colon cancer (61, 62). Sulindac also caused reduction of prostaglandin synthesis and regression of adenomatous polyps in 11 out of 15 patients with familial adenomatous polyposis (FAP), a condition in which many colorectal polyps develop spontaneously with eventual progression to tumors. That COX activity is involved in the process leading to colon cancer, is supported by the demonstration that COX-2 and not COX-1 is highly expressed in human and animal colon cancer cells as well as in human colorectal adenocarcinomas (63, 64). Further support for the close connection between COX-2 and colon cancer has come from studies in the mutant Apo mouse, which is a model of FAP in humans. The spontaneous development of intestinal polyposis in these mice was strongly reduced either by deletion of the COX-2 gene or by treatment with a highly selective COX-2 inhibitor (65—67). Nimesulide also reduced the number and size of intestinal polyps in Min mice (68). Furthermore, the development of azoxymethane-induced colon tumours over a year was inhibited in celecoxib-fed rats (69). A clinical trial of celecoxib in patients with FAP (70) has shown a positive reduction in polyps and this indication for the drug has been allowed by the FDA.

Alzheimer's disease

The connection between COX and Alzheimer's disease has been based mostly on epidemiology, because of the lack of an animal model of the disease. A number of studies have shown a significantly reduced odds ratio for Alzheimer's disease in those taking NSAIDs as anti-inflammatory therapy (71—73). The Baltimore Longitudinal Study of Ageing (74), with 1686 participants showed that the risk of developing Alzheimer's disease is reduced among users of NSAIDs, especially those who have taken the medications for 2 years or more. No decreased risk was evident with acetaminophen or aspirin use. However, aspirin was probably taken in a dose too low to have an anti-inflammatory effect. The protective effect of NSAIDs is consistent with evidence of inflammatory activity in the pathophysiology of Alzheimer's disease. There is a strong interest in COX-2 in Alzheimer's disease, and expression of COX-2 has been shown in the frontal cortex of brain from Alzheimer's patients (75).

CONCLUSIONS

Selective inhibitors of COX-2 clearly provide important advances in the therapy of inflammation. Conventional NSAIDs are associated with gastro-intestinal side effects, which include ulceration of the stomach,
sometimes with subsequent perforation and deaths estimated at several thousand a year in the USA alone. Selective COX-2 inhibitors have substantially reduced side effects on the stomach. Already, the published extensive clinical results with meloxicam show this improved safety and tolerability. The clinical trial results with rofecoxib or celecoxib are just as dramatic. In addition to their beneficial actions in inflammatory diseases, these drugs may be useful in the future for the prevention of colon cancer, Alzheimer’s disease or premature labour.

Finally, the suppression of prostacyclin release from endothelial cells by “specific” COX-2 inhibitors (34) suggests the possibility of interference with the cardio-vascular system. However, we have been using COX-2 inhibitors for many years, because this is how the NSAIDs produce their therapeutic effects. Thus, the “selective COX-2” inhibitors will do nothing different to prostacyclin production than a conventional NSAID, although the prostacyclin thromboxane balance may be changed because of their lack of effect on platelet COX-1.

New side effects of the selective COX-2 inhibitors, if any, may arise from the fact that they cross the blood-brain barrier, far more easily than do the conventional carboxy-acid NSAIDs.

REFERENCES


17. Tsai AI, Sanduja R and Wu KK. Evidence for two pools of prostaglandin H synthase in human endothelial cells. 7th International Conference on Prostaglandins and related compounds. Florence, Italy. 1990 Abstract No 41.


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