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EXPERIMENTAL STUDY ON VASOCONSTRICTION AND INFLAMMATION: ROLE OF LIPOXYGENASE PRODUCTS

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ABSTRACT

A sufficient blood supply is vital for the physiological functioning of the brain, and a lack of cerebral blood flow leads to neurodegenerative diseases. Immune cells defend the brain system, but they can be protagonists of inflammatory processes in pathological cases. Vasoconstriction and inflammation are pathological elements of the brain. Here, we study the pathogenetic mechanisms of vasoconstriction and inflammation in a rodent model in relation to arachidonic acid compounds. Experimentally induced inflammation was treated with various lipoxygenase and cyclooxygenase inhibitors. Lipoxygenase was inhibited by specific compounds but not by cyclooxygenase inhibitors, although both were anti-inflammatory.

KEYWORDS: *lipoxygenase, arachidonic acid, leukotriene, hydroxyeicosatetraenoic acid, leukocyte, SRS-A*

INTRODUCTION

The brain requires a sufficient blood supply for physiological functioning. A deficiency of cerebral blood flow leads to neurodegenerative diseases (1). In recent years, the importance of lipoxygenase as a key enzyme in inflammatory reactions and allergic processes has been increasingly recognized. The metabolites of arachidonic acid generated after stimulation of the lipoxygenase pathway (2), e.g., the hydroxy fatty acids and the leukotrienes, can regulate both cellular and humoral components of inflammatory and allergic reactions (6). In the first figure, some activities of lipoxygenase products are shown (Fig.1).

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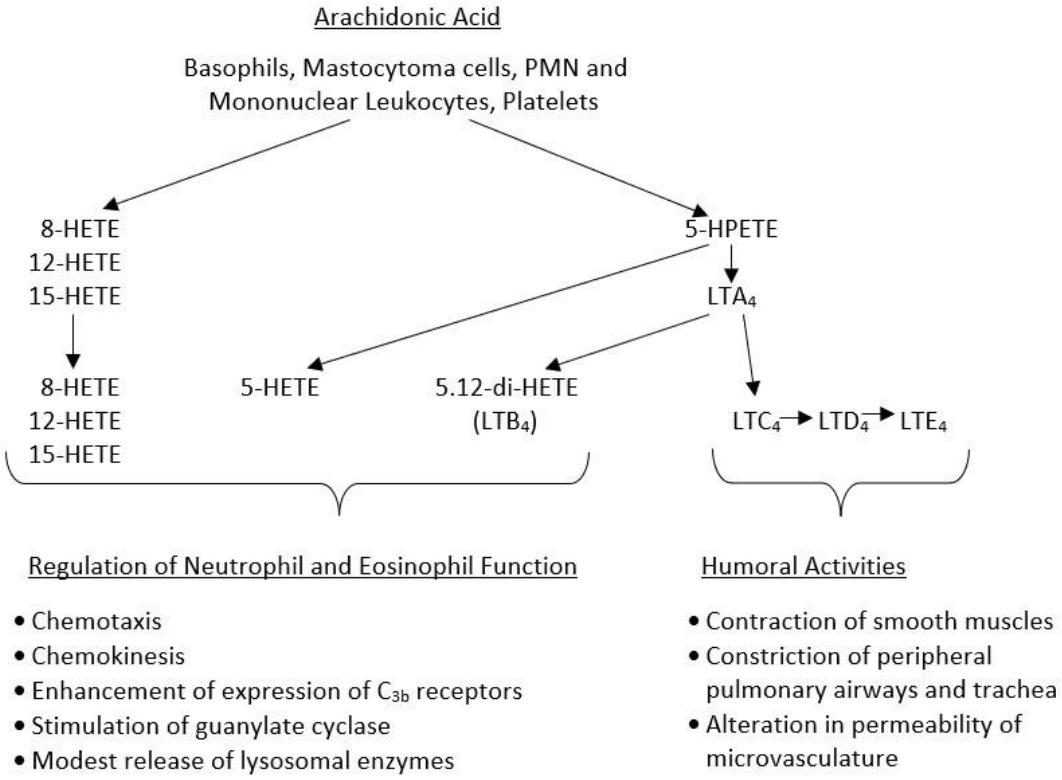

Fig. 1. *Generation and biological properties of products of arachidonic acid. HPETE = hydroperoxyeicosatetraenoic acid, LT = leukotriene.*

The hydroxyeicosatetraenoic acids (HETEs) possess leukotactic and leukinetic activities in which the potencies differ from the kind of metabolite (3). Leukotriene B4 is the most potent chemotactic factor for polymorphonuclear leukocytes. Vascular effects of this metabolite have also been observed due to increased permeability (4).

The stimulation of the leukocyte migration and the vascular effects are reactions which are important for the responses during inflammatory processes (5). Remarkably, some lipoxygenase products have modulating influences on the immune system (6). Thus, the expression of C3b receptors is enhanced, and various lipoxygenase products induce T-suppressor cells. Another important action for inflammatory and allergic reactions is the release of lysosomal enzymes (7).

The mechanism of action of the lipoxygenase products is possibly mediated via stimulating the guanylate cyclase and increasing the cGMP level (8). The leukotrienes C4, D4, and E4, components of SRS-A, are mediators released mainly during allergic reactions while also having been reported to play a role in inflammation (9). They have humoral activities and contract specific smooth muscles, e.g., ileum, peripheral pulmonary airways, and trachea. In addition, these compounds increase vascular permeability, leading to plasma exudation (10,11).

Due to the multiple actions of the lipoxygenase products, potent lipoxygenase inhibitors with favorable pharmacokinetics should, therefore, influence inflammatory and allergic reactions. Combining such agents with antagonists of other mediators of inflammation and allergic reactions could lead to more successful treatment (12).

The study of the pharmacological properties of drugs influencing inflammation and allergy is complicated by several different mediators that can be released during these reactions (13). It is further complicated by the fact that some of these mediators can have opposite effects and by the variety of animal models employed, some of which may not simulate the pathological situation in man (14).

An optimal test hierarchy should include investigating drugs' influence on the isolated enzyme, and such models in which mediators synthesized by the lipoxygenase are important for the pathological process (15).

We tested some compounds (Table I) to inhibit the reticulocyte lipoxygenase from rabbit and soybean lipoxygenase 1.

Table I. *Investigated compounds.*

BW 755 C	Indomethacin
Nordihydroguaiaretic acid (NDGA)	Acetylsalicylic acid (ASA)
Salicylhydroxamic acid (SHAM)	Phenylbutazone
FPL 55712	Noradrenaline
Propyl gallate	Adrenaline
Benoxaprofen	Isoprenaline
Diclofenac -Na	H 252
M 516	

The reticulocyte lipoxygenase converts arachidonic acid into the major product 15-HETE, and 12-HETE as the minor component (16), and the soybean lipoxygenase-1 forms 15-HPETE from arachidonic acid (17).

The anti-inflammatory and antiallergic activities of the compounds have been investigated by using the carrageenin oedema of rat paw as a model of acute inflammation, the adjuvant arthritis of the rat as a model of chronic inflammation, and the active anaphylactic oedema of rat paw as well as the active cutaneous anaphylaxis of the rat, as allergic models (18).

MATERIALS AND METHODS

Reticulocyte lipoxygenase was isolated from the defibrinated blood of rabbits and made anaemic by repeated bleeding or treatment with phenylhydrazine. The purification procedure involved ammonium sulfate precipitation, anion exchange chromatography on DEAE-Sephadex A-50, and isoelectric focusing in ampholine, pH 5 to 7.

Soybean lipoxygenase was obtained from Boehringer Mannheim GmbH (FRG). The activity of the lipoxygenase was measured polarographically through an oxygen electrode. The reaction mixture contained 0.53 mM linoleic acid, 0.2% sodium cholate, and 5% ethanol in 0.1 M potassium phosphate buffer, pH 7.4 and 9.0, respectively, and the final volume was 2.0 ml. Inhibitors were preincubated with the enzyme for 10 min. The assays were performed at 20° C.

Carrageenin oedema of the rat paw was induced in female Wistar rats by subplantar injection of 0.1 ml of a 1% carrageenin solution. The drugs were given orally and simultaneously with the carrageenin injection. Adjuvant arthritis was induced in female Wistar rats by subplantar injection of 0.1 ml Freund's complete adjuvant (0.5% suspension of heat-killed mycobacterium. in paraffinic perliquidum). The drugs were given orally once a day.

For producing the anaphylactic reactions, female Wistar rats were sensitized by i.m. injecting 0.2 ml of a 0.5% solution of bovine serum albumin in Bordetella pertussis vaccine containing $2x10^{10}$ bacteria. Two weeks later, the animals were challenged by subplantar injection of 500 μg bovine serum albumin in 0.1 ml 0.9% NaCl solution for producing the active anaphylactic oedema and by intracutaneous injection of 0.5 μg bovine serum albumin in 0.05 ml 0.9% NaCl solution for performing active cutaneous anaphylaxis, respectively. The drugs were administered orally 1 hour before the provocation of the edema and intraperitoneally 20 minutes before the cutaneous anaphylaxis, respectively.

RESULTS AND DISCUSSION

Generally, soybean lipoxygenase seems more insensitive to the inhibitors than reticulocyte lipoxygenase (19) (Table II). Therefore, the most potent inhibitors are BW 755 C, inhibiting cyclooxygenase (15), NDGA, SHAM, and propyl gallate.

Table II. *Inhibition of the activity of the reticulocyte lipoxygenase and soybean lipoxygenase-1.*

Compound	Inhibitory potency Reticulocyte lipoxygenase	Soybean lipoxygenase-1
BW 755 C	$IC_{50}= 3x10^{-6}$ M/l	$IC_{50}= 1.7x10^{-4}$ M/l
NDGA	$IC_{50}= 4x10^{-6}$ M/l	$IC_{50}= 2x10^{-5}$ M/l
SHAM	$IC_{50}= 5.6x10^{-5}$ M/l	$IC_{50}= 4.5x10^{-4}$ M/l
FPL 55712	$IC_{50}= 4x10^{-4}$ M/l	10^{-3}M/l 39% inhib.
Propyl gallate	$IC_{50}= 4x10^{-5}$ M/l	$IC_{50}= 9.4x10^{-5}$ M/l
Benoxaprofen	10^{-3}M/l 50% inhib.	10^{-3}M/l no inhib.
Diclofenac	10^{-3}M/l 53% inhib.	10^{-3}M/l no inhib.
Indomethacin	10^{-3}M/l 51% inhib.	10^{-3}M/l no inhib.
ASA	10^{-3}M/l no inhib.	10^{-3}M/l no inhib.
Phenylbutazone	10^{-3}M/l no inhib.	10^{-3}M/l no inhib.
Adrenaline	10^{-3}M/l no inhib.	10^{-3}M/l no inhib.
Noradrenaline	10^{-3}M/l 27% inhib.	10^{-3}M/l no inhib.
Isoprenaline	10^{-3}M/l no inhib.	10^{-3}M/l no inhib.
H 252	10^{-3}M/l no inhib.	10^{-3}M/l no inhib.
M 516	$IC_{50}= 6x10^{-5}$ M/l	$IC_{50}= 5.4x10^{-4}$ M/l

Remarkably, the SRS-A antagonist FPL 55712 inhibits the lipoxygenase activity, too and does not only display its antiallergic activity by antagonising the actions of the leukotrienes at the receptor site. The antiallergic and anti-inflammatory agent benoxaprofen blocks the lipoxygenase activity very weakly in our system (20). This drug selectively inhibits the 5-lipoxygenase, which is responsible for the biosynthesis of the precursors of LTC_4, D_4, and E_4 (21).

Of the anti-inflammatory drugs, only diclofenac and indomethacin exert weak inhibitory activities against the reticulocyte lipoxygenase. Catecholamines inhibit inflammatory responses (22) but do not affect lipoxygenase activity. The compound M 516 is shown to be a strong, selective inhibitor of lipoxygenase because it does not inhibit cyclooxygenase (23).

In investigating the compounds in the *in vivo* models, BW 755 C inhibits the carrageenin oedema after systemic and local administration (Tables III, IV).

Table III. *Inhibition of the carrageenin edema of rat paw ($^{++}$);($^{+}$) significantly different from control group ($p<0.01$; $p<0.05$) according to Student's t-test.*

Compound Dose/Administr.	% Inhibition 0.5 h	1 h	2 h	3 h	4 h	5 h
BW 755 C 50 mg/kg p.o.		35^{++}	37^{++}	62^{++}	62^{++}	60^{++}
BW 755 C 1 mg local.		29^{+}	33^{+}	21^{+}	-20	-10
NDGA 200 mg/kg p.o.		0	0	0	0	0
SHAM 200 mg/kg p.o.		9	0	0	0	7
FPL 55712 0.2 mg local.	45^{++}	25	-37	-4	-7	5
Propyl gallate 250 mg/kg p.o.				39^{++}		19
Propyl gallate 1 mg local.			0	20		9
Benoxaprofen 20 mg/kg p.o.		41^{+}	35^{+}	23	16	19
Diclofenac 2.5 mg/kg p.o.				49^{++}		22
Indomethacin 0.1 mg/kg p.o.			35^{++}	41^{++}		34^{++}
Indomethacin 0.1 mg local.			38^{++}	49^{++}		45^{++}
ASA 250 mg/kg p.o.				53^{++}	57^{++}	53^{++}
Phenylbutazone 100 mg/kg p.o.				57^{++}	62^{++}	54^{++}
Phenylbutazone 0.5 mg local.		38^{+}	57^{+}	45^{+}	38^{+}	22^{+}
Adrenaline 0.01 ug local.	49^{++}	54^{++}	7	9		17
Noradrenaline 0.01 ug local.	27^{+}	18	13	4		6
Isoprenaline 0.01 ug local.	29^{+}	36^{+}	10	7		7
H 252 65 mg/kg p.o.			58^{++}	50^{++}	26	7
M 516 68 mg/kg p.o.			41^{+}	45^{+}	45^{+}	34
M 516 1 mg local.		37^{+}	49^{+}	0	-18	2

Table IV. *Inhibition of the adjuvant arthritis of rat ($^{++}$);($^{+}$) significantly different from the control group ($p<0.01$; $p<0.05$) according to Student's t-test.*

Compound Dose/Administr.	% Inhibition 2 d	4 d	16 d
BW 755 C 50 mg/kg p.o.	23^{+}	20^{+}	
Propyl gallate 250 mg/kg p.o.	2	6	0
Benoxaprofen 20 mg/kg p.o.	33^{+}	35^{+}	
Diclofenac 2.5 mg/kg p.o.	41^{++}	51^{++}	52^{++}
Indomethacin 1 mg/kg p.o.	25^{+}	42^{++}	14^{+}
ASA 250 mg/kg p.o.		44^{++}	49^{++}
Phenylbutazone 100 mg/kg p.o.		52^{++}	59^{++}
Adrenaline 0.25 mg/kg s.c.		9	0
M 516 68 mg/kg p.o.	0	0	

Strong inhibitory properties on the anaphylactic models are also evident, as shown in Tables V and VI (Tables V, VI). These results conclude that there is the best correlation between *in vitro* and *in vivo* activities. However, the *in vivo* effects can also be caused by inhibiting cyclooxygenase.

Table V. *Inhibition of the active anaphylactic oedema of rat paw ($^{+}$); significantly different from the control group ($p<0.05$) according to Student's t-test.*

Compound Dose/Administr.	% Inhibition 0.5 h	1 h	2 h	3 h	4 h	5 h
BW 755 C 50 mg/kg p.o.	27	29	40^{+}	38^{+}	34^{+}	41^{+}
Benoxaprofen 50 mg/kg p.o.	42^{+}	47^{+}	51^{+}	38^{+}	54^{+}	52^{+}
Diclofenac 3 mg/kg p.o.	-7	0	15	20	14	23
Indomethacin 5 mg/kg p.o.	14	16	25	20	25	28
ASA 200 mg/kg p.o.	12	32	35^{+}	27	52^{+}	31
Phenylbutazone 30 mg/kg p.o.	18	7	2	18	5	27
H 252 100 mg/kg p.o.	14	27	13	4	6	8
M 516 100 mg/kg p.o.	6	3	24	2	-3	1

Table VI. *Inhibition of the active cutaneous anaphylaxis of the rat (+); significantly different from the control group (p<0.05) according to Student's t-test.*

Compound Dose/Administr.	% Inhibition
BW 755 C 50 mg/kg i.p.	43+
Benoxaprofen 50 mg/kg i.p.	48+
Diclofenac 5 mg/kg i.p.	6
Indomethacin 5 mg/kg i.p.	22
Phenylbutazone 30 mg/kg i.p.	12
ASA 200 mg/kg i.p.	27
H 252 100 mg/kg i.p.	22
M 516 100 mg/kg i.p.	18

The antioxidant propyl gallate inhibits carrageenin oedema only after oral administration at relatively high doses. Therefore, it does not affect adjuvant arthritis (24).

SHAM and NDGA show no influence on carrageenin oedema. Despite the suitable inhibitory activities on the lipoxygenase, there is no correlation with the *in vivo* properties of the propyl gallate, SHAM, and NDGA compounds. The cause of this infectivity could be insufficient absorption with ineffective serum and tissue concentration. The SRS-A antagonist FPL 55712 decreases the swelling of the carrageenin oedema in the first hour, suggesting that the C_4, D_4, and E_4 leukotrienes or other lipoxygenase products are released (25).

As an inhibitor of the 5-lipoxygenase, benoxaprofen acts dose-dependently on carrageenin oedema and adjuvant arthritis; these results are also obtained in the anaphylactic models. Acid nonsteroidal anti-inflammatory agents have the strongest inhibitory activities on carrageenin oedema and adjuvant arthritis and are cyclooxygenase inhibitors (26), while acetylsalicylic acid also significantly inhibits the anaphylactic paw oedema but not cutaneous anaphylaxis. Carrageenin oedema is inhibited by the catecholamines after local administration; however, inhibition of adjuvant arthritis could not be found (27).

The compounds H 252, which does not inhibit lipoxygenase activity, and M516, which inhibits lipoxygenase, show moderate dose-dependent inhibitory activities in carrageenin oedema. However, both drugs have no statistically significant influence on the anaphylactic models. The compound M 516 is also ineffective in the primary phase of adjuvant arthritis.

It can be stated that there are no satisfactory correlations between the inhibition of the 15- lipoxygenase and *in vivo* activities. The cause could be the subordinate importance of the products of these lipoxygenases in our *in vivo* models and/or insufficient substance concentrations *in vivo*. The best correlation is obtained with BW 755 C, which influences all *in vivo* models but is also a potent cyclooxygenase inhibitor.

CONCLUSIONS

The importance of lipoxygenase in inflammatory and allergic reactions is established. It oxidises multiple unsaturated fatty acids into many biologically active compounds, such as hydroxyeicosatetraenoic acids and leukotrienes. Therefore, the inhibitors of the lipoxygenase may be possible anti-inflammatory and antiallergic drugs (28).

Here, some compounds were tested for their activity against the isolated lipoxygenase from rabbit reticulocytes and soybean and in four *in vivo* models. Again, conventional anti-inflammatory drugs with high potency are tested as a standard.

The most potent lipoxygenase inhibitors were BW 755 C, NDGA, SHAM, and the compound M 516, which showed anti-inflammatory and antiallergic activities in some *in vivo* models (29). On the other hand, the potent cyclooxygenase inhibitors' anti-inflammatory agents had no inhibitory action on the lipoxygenase, but they strongly blocked the inflammatory reactions *in vivo* (30).

Conflict of interest

The authors declare that they have no conflict of interest.

REFERENCES

1. Duvall MG, Levy BD. DHA- and EPA-derived resolvins, protectins, and maresins in airway inflammation. *European Journal of Pharmacology*. 2016;785:144-155. doi:https://doi.org/10.1016/j.ejphar.2015.11.001
2. Powell WS, Rokach J. Biosynthesis, biological effects, and receptors of hydroxyeicosatetraenoic acids (HETEs) andoxoeicosatetraenoic acids (oxo-ETEs) derived from arachidonic acid. *Biochimica et Biophysica Acta (BBA) - Molecular and Cell Biology of Lipids*. 2015;1851(4):340-355. doi:https://doi.org/10.1016/j.bbalip.2014.10.008
3. Bitencourt CS, Bessi VL, Huynh DN, et al. Cooperative role of endogenous leucotrienes and platelet-activating factor in ischaemia–reperfusion-mediated tissue injury. *Journal of Cellular and Molecular Medicine*. 2013;17(12):1554-1565. doi:https://doi.org/10.1111/jcmm.12118
4. Byrum RS, Goulet JL, Snouwaert JN, Griffiths RJ, Koller BH. Determination of the Contribution of Cysteinyl Leukotrienes and Leukotriene B4 in Acute Inflammatory Responses Using 5-Lipoxygenase- and Leukotriene A4 Hydrolase-Deficient Mice. *The Journal of Immunology*. 1999;163(12):6810-6819. doi:https://doi.org/10.4049/jimmunol.163.12.6810
5. Muller WA. Getting Leukocytes to the Site of Inflammation. *Veterinary Pathology*. 2013;50(1):7-22. doi:https://doi.org/10.1177/0300985812469883
6. Catanzaro M, Corsini E, Rosini M, Racchi M, Lanni C. Immunomodulators Inspired by Nature: A Review on Curcumin and Echinacea. *Molecules*. 2018;23(11):2778. doi:https://doi.org/10.3390/molecules23112778
7. Ribeiro D, Freitas M, Lima JLFC, Fernandes E. Proinflammatory Pathways: The Modulation by Flavonoids. *Medicinal Research Reviews*. 2015;35(5):877-936. doi:https://doi.org/10.1002/med.21347
8. Dobrian AD, Lieb DC, Cole BK, Taylor-Fishwick DA, Chakrabarti SK, Nadler JL. Functional and pathological roles of the 12- and 15-lipoxygenases. *Progress in Lipid Research*. 2011;50(1):115-131. doi:https://doi.org/10.1016/j.plipres.2010.10.005
9. Bäck M. Leukotriene Signaling in Atherosclerosis and Ischemia. *Cardiovascular Drugs and Therapy*. 2008;23(1):41-48. doi:https://doi.org/10.1007/s10557-008-6140-9
10. Claesson-Welsh L. Vascular permeability—the essentials. *Upsala Journal of Medical Sciences*. 2015;120(3):135-143. doi:https://doi.org/10.3109/03009734.2015.1064501
11. Nagy JA, Benjamin L, Zeng H, Dvorak AM, Dvorak HF. Vascular permeability, vascular hyperpermeability and angiogenesis. *Angiogenesis*. 2008;11(2):109-119. doi:https://doi.org/10.1007/s10456-008-9099-z
12. Sala A, Proschak E, Steinhilber D, Rovati GE. Two-pronged approach to anti-inflammatory therapy through the modulation of the arachidonic acid cascade. *Biochemical Pharmacology*. 2018;158:161-173. doi:https://doi.org/10.1016/j.bcp.2018.10.007
13. Gómez E, Torres MJ, Mayorga C, Blanca M. Immunologic Evaluation of Drug Allergy. *Allergy, Asthma & Immunology Research*. 2012;4(5):251. doi:https://doi.org/10.4168/aair.2012.4.5.251
14. Gregory NS, Harris AL, Robinson CR, Dougherty PM, Fuchs PN, Sluka KA. An Overview of Animal Models of Pain: Disease Models and Outcome Measures. *The Journal of Pain*. 2013;14(11):1255-1269. doi:https://doi.org/10.1016/j.jpain.2013.06.008
15. Meirer K, Steinhilber D, Proschak E. Inhibitors of the Arachidonic Acid Cascade: Interfering with Multiple Pathways. *Basic & Clinical Pharmacology & Toxicology*. 2013;114(1):83-91. doi:https://doi.org/10.1111/bcpt.12134
16. Kim BJ, Shin KC, Oh DK. Enzymatic Production of 15-Hydroxyeicosatetraenoic Acid from Arachidonic Acid by Using Soybean Lipoxygenase. *Journal of Microbiology and Biotechnology*. 2014;24(3):359-362. doi:https://doi.org/10.4014/jmb.1310.10054
17. Chawengsub Y, Gauthier KM, Campbell WB. Role of arachidonic acid lipoxygenase metabolites in the regulation of vascular tone. *American Journal of Physiology-Heart and Circulatory Physiology*. 2009;297(2):H495-H507. doi:https://doi.org/10.1152/ajpheart.00349.2009
18. Fehrenbacher JC, Vasko MR, Duarte DB. Models of Inflammation: Carrageenan- or Complete Freund's Adjuvant-Induced Edema and Hypersensitivity in the Rat. *Current protocols in pharmacology / editorial board, SJ Enna (editor-in-chief) [et al]*. 2012;0 5:Unit5.4. doi:https://doi.org/10.1002/0471141755.ph0504s56

19. Youn B, Sellhorn GE, Mirchel RJ, Gaffney BJ, Grimes HD, Kang C. Crystal structures of vegetative soybean lipoxygenase VLX-B and VLX-D, and comparisons with seed isoforms LOX-1 and LOX-3. *Proteins: Structure, Function, and Bioinformatics*. 2006;65(4):1008-1020. doi:https://doi.org/10.1002/prot.21182
20. Dawson W, Boot JR, Harvey J, Walker JR. The pharmacology of benoxaprofen with particular to effects on lipoxygenase product formation. *European Journal of Rheumatology and Inflammation*. 1982;5(2):61-68.
21. Kobayashi I, Hamasaki Y, Sato R, et al. Saiboku-To, a herbal extract mixture, selectively inhibits 5-lipoxygenase activity in leukotriene synthesis in rat basophilic leukemia-1 cells. *Journal of Ethnopharmacology*. 1995;48(1):33-41. doi:https://doi.org/10.1016/0378-8741(95)01280-q
22. Gan TJ. Diclofenac: an update on its mechanism of action and safety profile. *Current Medical Research and Opinion*. 2010;26(7):1715-1731. doi:https://doi.org/10.1185/03007995.2010.486301
23. Rao CV, Janakiram NB, Mohammed A. Lipoxygenase and Cyclooxygenase Pathways and Colorectal Cancer Prevention. *Current Colorectal Cancer Reports*. 2012;8(4):316-324. doi:https://doi.org/10.1007/s11888-012-0146-1
24. Woode E, Ainooson G, Amo-Barimah A, Boakye-Gyasi E, Abotsi Wonder MK. Antiarthritic and antioxidant effects of the leaf extract of Ficus exasperate P. Beauv. (Moraceae). *Pharmacognosy Research*. 2010;2(2):89. doi:https://doi.org/10.4103/0974-8490.62958
25. Hsu HC, Lin WC, Chang PJ, Hong CZ, Chen CH. Propyl gallate inhibits TPA-induced inflammation via the nuclear factor-κB pathway in human THP-1 monocytes. *Experimental and Therapeutic Medicine*. 2013;5(3):964-968. doi:https://doi.org/10.3892/etm.2013.896
26. Hiermann A, Schramm HW, Laufer S. Anti-inflammatory activity of myricetin-3-O-β-D-glucuronide and related compounds. *Inflammation Research*. 1998;47(11):421-427. doi:https://doi.org/10.1007/s000110050355
27. Poh J, Knowles S. Safety of 5-Aminosalicylic Acid Derivatives in Patients with Sensitivity to Acetylsalicylic Acid and Nonsteroidal Anti-inflammatory Drugs. *The Canadian Journal of Hospital Pharmacy*. 2014;67(1). doi:https://doi.org/10.4212/cjhp.v67i1.1318
28. Wisastra R, Dekker F. Inflammation, Cancer and Oxidative Lipoxygenase Activity are Intimately Linked. *Cancers*. 2014;6(3):1500-1521. doi:https://doi.org/10.3390/cancers6031500
29. Batt DG. 1 5-Lipoxygenase Inhibitors and their Anti-inflammatory Activities. *Progress in Medicinal Chemistry*. 1992;29:1-63.doi:https://doi.org/10.1016/s0079-6468(08)70004-3
30. Zarghi A, Arfaei S. Selective COX-2 Inhibitors: A Review of Their Structure-Activity Relationships. *Iranian journal of pharmaceutical research : IJPR*. 2011;10(4):655-683.