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GCP 28 238, a new potent nonsteroidal antiinflammatory agent: Its relation to arachidonic acid metabolism

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The highly potent nonsteroidal antiinflammatory compound CGP 28 238 (6-(2,4-difluorophenoxy)-5-methyl-sulfonylamino-1-indanone) does not inhibit prostaglandin (PG) synthase preparations of, e.g., sheep seminal vesicles, although it is a potent inhibitor of PG synthesis in cellular systems [1, 2]. We found that CGP 28 238 even antagonizes PG synthase inhibition caused by aspirin or indomethacin in sheep seminal vesicle (SSV) preparations. Furthermore, CGP 28 238 does not influence the incorporation of archidonic acid into lysophospholipids of rat platelets, neither *in vitro* nor *ex vivo* after oral administeration of the substance.

Material and methods

The activity of PG synthase in microsomal preparations of SSV was measured by determination of oxygen consumption in using an oxygen-sensitive electrode.

The incorporation of arachidonic acid into lysophospholipids of rat platelets was determined by using a technique described in detail elsewhere [3].

Animals used: female rats of an outbred Wistar strain (Wist: Barby), b.w. 270–320 g.

Substances employed: 1-^{14}C-arachidonic acid (60 mCi/mmol; Amersham International), aspirin (VEB Chemisch-pharmazeutisches Werk Oranienburg); CGP 28 238 (CIBA-GEIGY AG, Basel); diclofenac-Na (CIBA-GEIGY AG, Basel); indomethacin (Merck Sharp & Dohme, Rahway); Thiomersal (VEB Berlin-Chemie, Berlin). The determination of radioactively-labelled phosphatidylcholine was performed by using the liquid scintillation technique (PL 4540 Liquid Scintillations Counter, Phillips, UK).

Results and discussion

As shown in the Table, the 100% PG synthase-inhibiting activity by aspirin as well as by indomethacin, was reduced after preincubation of the SSV microsomal preparations with CGP 28 238 in increasing concentrations. At a concentration of 10^{-3} mol $\times 1^{-1}$, CGP 28 238 caused a weak PG synthase inhibition itself (21%). Therefore, the effect of the classical nonsteroids aspirin and indomethacin, cannot be completely abolished. The decrease by CGP 28 238 of the indomethacin-caused PG synthase inhibition seems to be due to a reversible competitive mechanism, since the CGP effect was reduced by increasing the indomethacin concentration up to 10^{-5} mol $\times 1^{-1}$ (data not shown). The mechanism of this interaction, still remains to be explained. In any case, this (pharmadynamic?) interaction could be a point for interpreting the underadditive antiinflammatory effects in combining CGP 28 238 and indomethacin during the primary phase of rat adjuvant arthritis [4]. Regarding the influence of CGP 28 238 on ^{14}C-arachidonic acid incorporation in lysophatidylcholine of rat platelets, we found no effect *in vitro* up to a CGP concentration of 10^{-3} mol $\times 1^{-1}$, and we also could not measure any influence *ex*

Table
Influence of CGP 28 238 on the PG synthase inhibiting activity of aspirin and indomethacin in SSV.

Concentration of CGP 28 238 mol × l^{-1}	PG synthase inhibition (%) by	
	Aspirin 10^{-3} mol × l^{-1}	Indomethacin 10^{-6} mol × l^{-1}
0	100	100
5×10^{-6}		95
1×10^{-5}	85	79
3×10^{-5}	69	63
7×10^{-5}	43	43
1×10^{-4}	33	28
1×10^{-3}	39	30

Inhibition of PG synthase activity by CGP 28 238 at a concentration of 10^{-3} mol × l^{-1}: 21%.

vivo after an oral CGP dose of 5 mg/kg. In contrast, aspirin (10^{-5} mol × l^{-1}) and diclofenac (10^{-4} mol × l^{-1}) caused a significant 20–25% increase of arachidonic acid incorporation in lysophospholipids *in vitro*, which is apparently due to PG synthase inhibition. As expected, an increased amount of labelled phosphatidylcholine could be found *ex vivo* after aspirin administration only (200 mg/kg p.o.) [3]. Diclofenac was inactive *in vivo*/*ex vivo* up to an oral dose of 10 mg/kg, which is apparently due to weak-binding affinity of diclofenac to the PG synthase. Thiomersal (10^{-4} mol × l^{-1}], an inhibitor of lysophospholipid acyltransferase [5], caused a significant 50% inhibition of arachidonic acid incorporation in platelet lysophospholipids *in vitro* [3].

To summarize, we may state that the potent nonsteroid antiinflammatory agent CGP 28 238 interacts with aspirin and indomethacin at the PG synthase of microsomal SSV preparations, causing a reduction of the PG synthase inhibition by these two classical nonsteroidal agents. This effect could explain *in vivo* interactions in inflammatory reactions. Furthermore, CGP 28 238 apparently does not influence the incorporation of arachidonic acid in lysophospholipids of rat platelets. The mode of CGP 28 238 action, which indeed inhibits the PG synthesis in inflamed tissues [1], has yet to be established, mainly its relation to arachidonic acid metabolism.

References

[1] I. Wiesenberg-Boettcher, A. Schweizer, J. R. Greene, K. Mueller, F. Maerki and J. Pfeilschifter, *The pharmacological profile of CGP 28 238, a novel highly potent anti-inflammatory compound.* Drugs Expl. Clin. Res. *XV,* 501–510 (1989).

[2] I. Wiesenberg-Boettcher, A. Schweizer, J .R. Greene, Y. Seltenmeyer and K. Mueller, *The pharmacological profile of CGP 28 238, a highly potent anti-inflammatory compound.* Agents and Actions, *26,* 240–242 (1989).

[3] M. Hentschel, *Methodische Untersuchungen zur Reacylierung von Lysophosphatidylcholin mit Arachidonsäure in unstimulierten Rattenthrombozyten sowie Einfluß von steroidalen und nichtsteroidalen Antiphlogistika.* Thesis, Halle 1989.

[4] R. Hirschelmann and G. Pöch. unpublished.

[5] M. Goppelt-Strübe, C. F. Körner, G. Hausmann, D. Gemsa and K. Resch, *Control of prostanoid synthesis. Role of reincorporation of released precursor fatty acids.* Prostaglandins *32,* 373–385 (1986).