Letters in Peptide Science, 2 (1995) 135–140
ESCOM

LIPS 075

New peptidomimetics in the chemistry of fibrinogen receptor antagonists

J. Gante*, H. Juraszyk, P. Raddatz, H. Wurziger, S. Bernotat-Danielowski, G. Melzer and F. Rippmann

Merck KGaA, Preclinical Pharmaceutical Research Department, Frankfurter Strasse 250, D-64271 Darmstadt, Germany

Received 8 September 1995
Accepted 2 November 1995

Keywords: RGD; Antithrombotics; GP IIb/IIIa

SUMMARY

RGD-peptidomimetics are currently being investigated as a class of potential antithrombotics that antagonize the fibrinogen receptor, GP IIb/IIIa, on the surface of platelets. These mimetics are expected to have decisive advantages – such as higher activity and specificity, oral bioavailability and longer duration of action – over known antithrombotics. For further optimization in this respect, novel peptidomimetic GP IIb/IIIa antagonists with an oxazolidinonemethyl central building block were synthesized. This building block proved to be very versatile as an 'anchor' for structurally different C-termini and was the starting point for highly efficient and orally active compounds.

INTRODUCTION

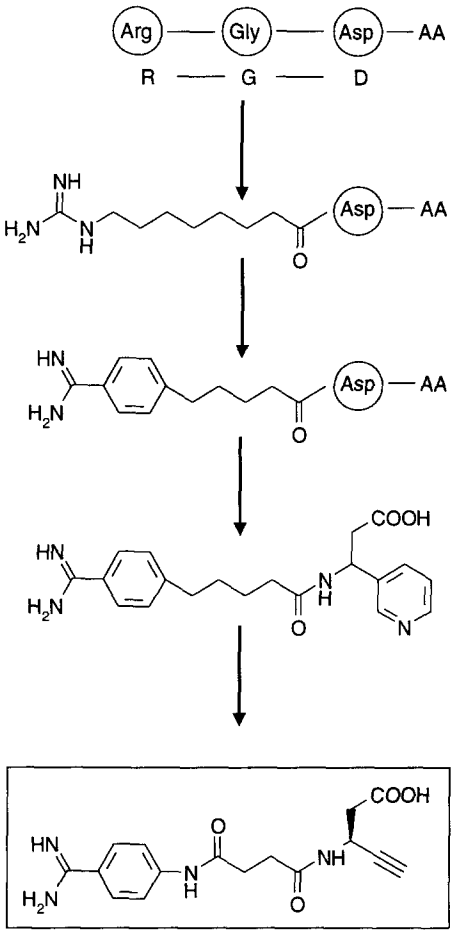
Analogues of the RGD sequence in certain bioactive peptides or proteins are, as so-called 'adhesion receptor antagonists', very important candidates as antithrombotics [1].

These potential pharmaceuticals are aimed to antagonize adhesion receptors of the GP IIb/IIIa type on blood platelets. These receptors are glycoproteins of the large family of 'integrins', which undergo binding to the blood protein fibrinogen as an essential step during the coagulation process. Blocking them by bioactive substances should interfere with and ameliorate pathological conditions such as thrombosis, unstable angina, acute myocardial infarction, thrombotic stroke and peripheral arterial occlusion [2]. The receptors are located on the surface of blood platelets (~ 50 000 per platelet), which play an important role in the coagulation process.

The GP IIb/IIIa receptor is a transmembrane heterodimer with binding sites for the attachment of fibrinogen. Fibrinogen itself has binding sites for the attachment to the platelet receptor, which are characterized by R-G-D (Arg-Gly-Asp) sequences within the protein chain. The multiple occurrence of the 'binding points' on both sides implies the formation of a tight platelet/fibrinogen network during the clotting process.

For the development of nonpeptidic adhesion

*To whom correspondence should be addressed.



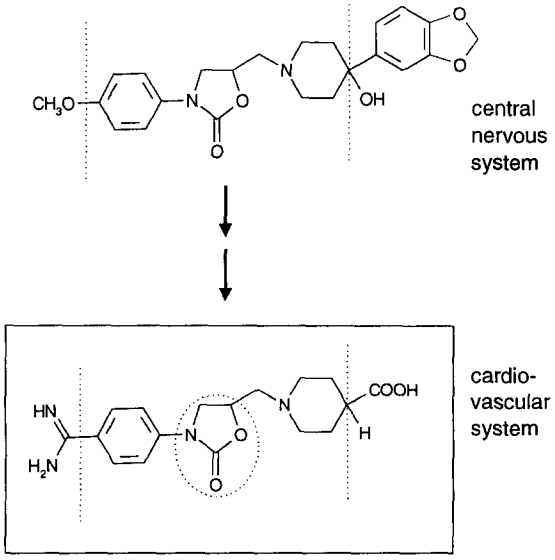
Scheme 1. Mimetization sequence from peptide to nonpeptide.

receptor antagonists, the binding RGD sequence of the fibrinogen is taken as a motif for low-molecular-weight inhibitors that should bind more strongly to the receptor in order to prevent fibrinogen itself from binding [3], thus inhibiting platelet aggregation. The inhibitors should also be specific for this integrin and, if possible, orally active for chronic treatment. Therefore, in the development of such compounds, there is a trend from fibrinogen-derived peptides via 'semi-mimetics' to low-molecular-weight 'complete' nonpeptides [1,4]. This can be demonstrated by a structural 'mimetization sequence' with an assembly of structures synthesized at different laboratories as an example (see Scheme 1).

As a first logical step, arginine and glycine of the fibrinogen -Arg-Gly-Asp- sequence are modified, so that only the arginine guanidino group and the glycine carbonyl group remain, with a simple aliphatic chain as 'spacer'.

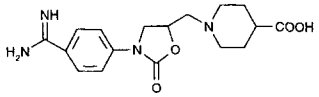
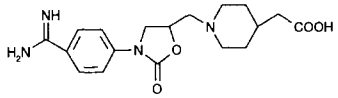
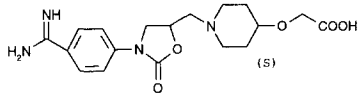
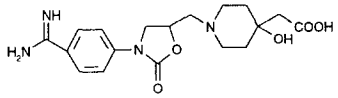
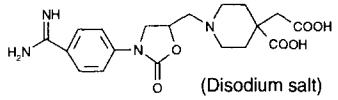
The next step on the way to 'less peptidic' compounds is the change from guanidino-alkyl to amidino-phenyl, which introduces a rigidizing element into the molecule and considerably enhances the binding strength. Then the aspartate residue is replaced with an unnatural β-amino acid residue. This leads to oral activity, which can be enhanced by further rigidization and introduction of an acetylenic β-amino acid, as shown in Scheme 1.

We want to report here the design, synthesis and biological activity of a new system with a rigidizing central oxazolidinonemethyl building block. This cardiovascularly active system is formally generated by a simple exchange of functional groups from a proven CNS-active (neuroleptic) Merck substance [5] (Scheme 2). Theoretical and computer modelling considerations, disclosing suitable conformational parameters of this building block, had encouraged us in this effort.



Scheme 2. From the central nervous system to the cardiovascular system by functional group exchange.

TABLE 1
STRUCTURE–ACTIVITY RELATIONSHIPS FOR A SERIES OF SUBSTITUTED PIPERIDINES

Piperidine	IC_{50} (μM)	
	Fibrinogen binding test	Collagen-induced platelet aggregation (PAT)
	0.0076	0.5
	0.122	5.0
 (S)	0.168	0.5
	0.036	>10
 (Disodium salt)	4.21	>10

The substitution on this piperidine building block proved to be variable over a wide range, giving highly active compounds. In Table 1 the structure–activity relationships of a series of substituted piperidines are listed. As can be seen, the binding strength is diminished with elongation of the chain, whereas a hydroxy group, but not an additional carboxy group, enhances the biological activity. On average, the antiaggregatory activity (PAT) of these molecules is moderate.

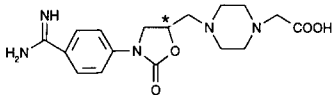
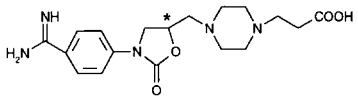
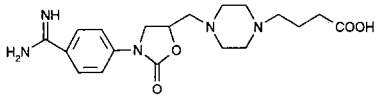
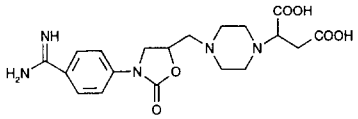
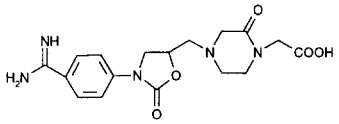
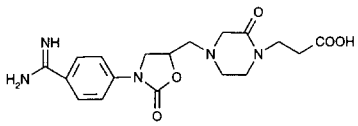
Exchanging piperidine by piperazine substituents gives another group of compounds with high biological activity (see Table 2). As can be seen, the optimum of the binding strength and antiplatelet aggregatory activity is found with the propionic acid, whereas further chain elongation gives a less active product. An interesting feature is the fact that in the acetic acid series the (*R*)-stereoisomer is the most active compound, whereas in the propionic acid series it is the (*S*)-isomer.

The synthesis of the new system is demonstrated with the example of a piperazine compound (see Scheme 3). Glycidol is reacted with 4-aminobenzonitrile to give the diol. This reacts with diethylcarbonate with ring closure to give the heterocyclic alcohol. Mesylation and reaction with the piperazino carboxylic acid derivative affords the basic system from which the functional groups of the final product are generated by successive reaction with hydroxylamine and hydrogenation.

Modification of the N-terminus to the corresponding aminomethyl and guanidinomethyl compounds by hydrogenation of the cyano group and successive guanylation of the amino groups led to compounds with decreased biological activity compared to the corresponding amidines (Table 3).

We also prepared compounds with piperidine and guanyl-piperidine functionalities instead of amidinophenyl. Here a strong increase in activity in the transition from the piperidine to the corre-

TABLE 2
STRUCTURE–ACTIVITY RELATIONSHIPS FOR A SERIES OF SUBSTITUTED PIPERAZINES

Piperazine	IC_{50} (μM)	
	Fibrinogen binding test	Collagen-induced platelet aggregation (PAT)
	0.124 (*RS*) 0.044 (*R*) 0.248 (*S*)	0.1 0.1 0.5
	0.0042 (*RS*) 0.008 (*R*) 0.0009 (*S*)	0.05 0.5 0.1
	1.18	0.05
	0.0077	0.05
	0.05	0.05
	0.0022	0.05

sponding guanidine was observed (Table 3). The starting materials are 1-benzyl-4-aminopiperidine and glycidol in a reaction sequence analogous to the synthesis of the amidinophenyl derivatives. The guanylation was performed with *N*-guanylpyrrazole [6].

That the heterocyclic mesylate is a versatile intermediate in the synthesis of potent GP IIb/IIIa antagonists could be shown by reaction with other nucleophiles, for instance with the sodium salts of the o-, m- and p-substituted hydroxyphenyl acetic acid esters to give (performing the usual reactions) the corresponding amidino carboxylic acids. The biological activities of these compounds are shown in Table 4. Interestingly, the para and ortho derivatives show high receptor affinities, while the meta compound is nearly inactive. The PAT value for the *o*-acetic acid is significantly higher than for the other two.

Most of the amidino compounds are orally active (in guinea pig) in the low mg/kg range. The most active compounds in this respect belong to the piperidine and piperazine series.

DISCUSSION AND CONCLUSIONS

The structure–activity relationships within the new system are as follows. In the piperidine series, the strongest receptor inhibition is shown by the 4-piperidine carboxylic acid derivative. With the piperazines, a slight enhancement of activity can be achieved. In this series the 4-piperazine propionic

TABLE 3
STRUCTURE–ACTIVITY RELATIONSHIPS FOR COMPOUNDS WITH A MODIFIED N-TERMINUS

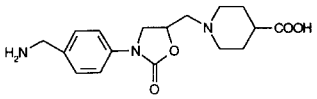
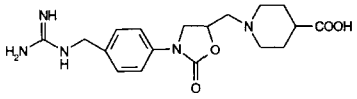
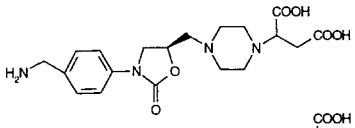
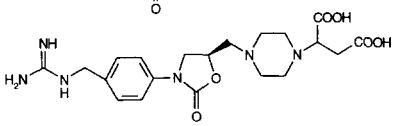
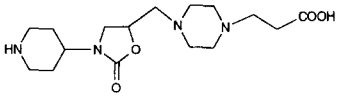
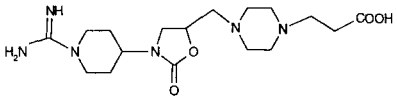
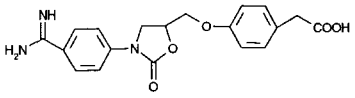
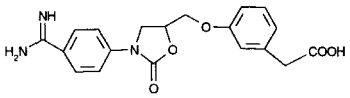
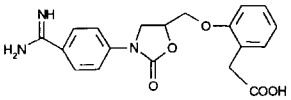
Variation of the N-terminus	IC_{50} (μM)	
	Fibrinogen binding test	Collagen-induced platelet aggregation (PAT)
	0.151	1.0
	0.062	10.0
	1.0	0.5
	1.37	1.0
	3.65	0.5
	0.18	0.5

TABLE 4
STRUCTURE–ACTIVITY RELATIONSHIPS FOR VARIOUS PHENYLACETIC ACIDS

Phenylacetic acid	IC_{50} (μM)	
	Fibrinogen binding test	Collagen-induced platelet aggregation (PAT)
	0.044	10.0
	183	> 10
	0.034	1.0

DOCKET ALARM

Explore Litigation Insights

Docket Alarm provides insights to develop a more informed litigation strategy and the peace of mind of knowing you're on top of things.

Real-Time Litigation Alerts

Keep your litigation team up-to-date with **real-time alerts** and advanced team management tools built for the enterprise, all while greatly reducing PACER spend.

Our comprehensive service means we can handle Federal, State, and Administrative courts across the country.

Advanced Docket Research

With over 230 million records, Docket Alarm's cloud-native docket research platform finds what other services can't. Coverage includes Federal, State, plus PTAB, TTAB, ITC and NLRB decisions, all in one place.

Identify arguments that have been successful in the past with full text, pinpoint searching. Link to case law cited within any court document via Fastcase.

Analytics At Your Fingertips

Learn what happened the last time a particular judge, opposing counsel or company faced cases similar to yours.

Advanced out-of-the-box PTAB and TTAB analytics are always at your fingertips.

API

Docket Alarm offers a powerful API (application programming interface) to developers that want to integrate case filings into their apps.

LAW FIRMS
Build custom dashboards for your attorneys and clients with live data direct from the court.

Automate many repetitive legal tasks like conflict checks, document management, and marketing.

FINANCIAL INSTITUTIONS
Litigation and bankruptcy checks for companies and debtors.

E-DISCOVERY AND LEGAL VENDORS
Sync your system to PACER to automate legal marketing.