iJRASET
International Journal For Research in Applied Science and Engineering Technology

INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Volume: 7 **Issue:** XI **Month of publication:** November 2019

DOI: http://doi.org/10.22214/ijraset.2019.11146

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International Journal for Research in Applied Science & Engineering Technology (IJRASET)
ISSN: 2321-9653; IC Value: 45.98; SJ Impact Factor: 7.177
Volume 7 Issue XI, Nov 2019- Available at www.ijraset.com

Docking of Cathepsin D with New Compounds

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Abstract: Cathepsin D is a lysosomal protein belongs to aspartic protease family. Over expression of this protein is observed in certain pathological conditions like apoptosis, Alzheimer's disease, Cancer, Atherosclerosis, Metastasis of breast cancer etc. Some of these pathological conditions can be cured by inhibiting this protein. Present study was focused on screening and docking of several chemical compounds like Imidazoles, Oxadiazoles, Pyrazoles, Pepstatins, Piperazines with cathepsin d protein. Crystal structure of cathepsin D (1LYW) was extracted from PDB. These ligands were docked into the active site of Cathepsin D to study their binding energy and interactions. From these docking studies it was noticed that the pepstatin is the strong and very potent inhibitor of cathepsin d with estimated Inhibition Constant [Ki] 1.26 nM and -14.70 Kcal/mol Estimated Binding Energy . Next best inhibitor was Norchlorcyclizine with Ki 53.23 nM and -9.92 Kcal/mol Estimated Binding Energy. Remaining all other tested compounds were found to be having moderate binding capacity with high Ki in micro molars range.
Key words : Cathepsin D, Docking, Cancer, Alzheimer disease, Metastasis.

I. INTRODUCTION

Cathepsin D mainly involves in the acidic degradation of lysosomal proteins. This protein is composed of two domains [6], which are separated by a large cleft. This cleft contains the active site that holds the substrate. Cathepsin D protein contains two catalytically active aspartic acid residues in its active site [Asp33 and Asp231] [7], which are contributed by each domain [6]. Cathepsin d active site can hold upto eight amino acids. Apart from these catalytically active aspartic acids, several amino acids from other regions in the protein forms hydrogen bonds with the substrate or inhibitors to strengthen the complex. By targeting Cathepsin d protein, we can study certain therapeutical areas like protein targeting [1], processing and presentation of antigens to MHC class II [2], muscular dystrophy [3], Alzheimer disease [4] degenerative brain diseases [5].

II. MATERIALS AND METHODS

1) *Ligands and Protein Preparation*: Crystal structure of cathepsin D (PDB: 1LYW) was collected from Protein Data Bank and the file was prepared by deleting all unwanted ligands and HETATM atoms. All the ligands structures were drawn using chemsketch tool. Both the ligands and protein energies were minimized and optimized. Ligands used for the study were belongs to the groups of Imidazoles, Oxadiazoles, Pyrazoles, Pepstatins, Piperazines.
2) *Molecular Docking*: AutoDock Tools 4.0 was used to perform docking. All the above processed ligands and protein structure were used for docking. Grid parameter files were generated in Auto grid. Grid dimensions on X, Y, Z were set to 60×60×60 Å. Then Autodock tool was used to dock all these ligands into the active site of cathepsin d protein. Cygwin was to generate the logarithmic files. Lamarckian Genetic Algorithm was used to process these files, which calculates the binding energies of the docked ligand and protein complex, binding orientations, hydrogen bonds between the ligand and protein amino acid molecules etc. Then by using UCSF-chimera three dimensional orientation of ligand protein complex and hydrogen bond interactions were visualized.

III. RESULTS AND DISCUSSION

Cathepsin d protein active site was docked with Imidazoles, Oxadiazoles, Pyrazoles, Pepstatins, Piperazines. Other than the active site amino acids, few aminoacids were also strongly interacted with the ligands. Clotrimazole formed strong interaction with the aminoacids ARG 245, LYS 249, ALA 253, GLY 252. Norchlorcyclizine interacted with ASN 38, GLN 113, LEU 83, LEU 99, PHE 115, ILE 76, VAL 114, VAL 147, VAL 144. 3-(3-Bromophenyl)-1H-pyrazole interacted with TYR 86, ASN 70, HIS 45, SER 61. 1,2,4-Oxadiazole-3-methylamine interacted with ASP 62, LYS 63, ASP 62. Pepstatin interacted with LEU 83, VAL 144, VAL 147, TYR 78, SER 143, ILE 134, TYR 10, SER 36, ILE 142, ARG 202, SER 315. Binding Energy (Kcal/mol), Inhibition Constant Ki, hydrogen bond interactions were studied and the results were incorporated in the below tables.

Table-1: Cathepsin d docking with Imidazoles
Calculation of Binding Energy, Inhibitory constant and of Imidazoles.

No	Ligand Name	Estimated Binding Energy Kcal/mol	Estimated Inhibition Constant Ki.	H-bonds
1.	Clotrimazole	-7.11	6.10 uM	5
2.	Isoconazole	-2.46	15.85 mM	6
3.	Bifonazole	-5.51	91.87 uM	4
4.	Econazole	-6.61	14.32 uM	4

The predicted binding energy of all the four Imidazoles were in between -2.46 Kcal/mol To -7.11 Kcal/mol and predicted inhibitory constant Ki values were in 6.10uM to 15.85 mM.

Table-II: Cathepsin d docking with Oxadiazoles
Calculation of Binding Energy, Inhibitory constant of oxadiazoles

No	Ligand Name	Estimated Binding Energy Kcal/mol	Estimated Inhibition Constant Ki.	H-bonds
1.	1,2,4-Oxadiazole-3-methylamine	-5.05	197.82 uM	6
2.	3-(chloromethyl)-5 methyl-1,2,4-oxadiazole	-3.19	4.62 mM	3
3.	3-Bromo-5-(bromomethyl)-1,2,4-oxadiazole	-3.59	2.32 mM	3
4.	5-phenyl-1,3,4-oxadiazole-2-thiol	-4.20	830.60 uM	6
5.	5-(4-Chlorophenyl)-1,3,4-oxadiazole-2-thiol	-4.56	457.05 uM	6
6.	5-methyl-1,3,4-oxadiazole-2-thiol	-3.10	5.32 mM	5

The predicted binding energy of all Oxadiazoles were in between -5.05 Kcal/mol to -3.10 Kcal/mol and predicted inhibitory constant Ki values were in between 197.82 uM to 5.32 mM .

Table-III: Cathepsin d docking with Piperazines:
Calculation of Binding Energy, Inhibitory constant of Piperazines

No	Ligand Name	Estimated Binding Energy Kcal/mol	Estimated Inhibition Constant Ki.	H-bonds
1.	1-(2-Chlorophenyl) piperazine	-5.26	138.53 uM	6
2.	1-(2-Furoyl) piperazine	-4.15	900.48 uM	5
3.	Aminoethyl piperazine	-4.90	255.71 uM	9
4.	Norchlorcyclizine	-9.92	53.23 nM	6
5.	1-Isopropyl piperazine	-4.33	667.89 uM	4
6.	1-(4-pyridyl) piperazine	-4.54	470.72 uM	4

The predicted binding energy of all the Piperazines were in between -4.15 Kcal/mol to -9.92 Kcal/mol and predicted inhibitory constant Ki values were in between 53.23 nM to 900.48 uM.

Table-IV: Cathepsin d docking with Pyrazoles
Calculation of Binding Energy, Inhibitory constant of pyrazole

No	Ligand Name	Estimated Binding Energy Kcal/mol	Estimated Inhibition Constant Ki.	H-bonds
1.	1H-Pyrazole-3-carbaldehyde	-3.24	4.22 mM	7
2.	1H-pyrazole-4-carbaldehyde	-3.20	4.50 mM	7
3.	2,3-dihydro-1H-imidazo(1,2-b)pyrazole or Imidazolepyrazole	-3.33	3.60 mM	6
4.	3-(3-Bromophenyl)-1H-pyrazole	-4.69	362.56 uM	6
5.	3-(4-Fluorophenyl)-5 (Methylthio)Pyrazole	-4.17	883.18 uM	5
6.	4-carboxypyrazole	-3.56	2.45 mM	7

The predicted binding energy of all the nine pyrazole were in between -3.20 Kcal/mol to -4.69 Kcal/mol and predicted inhibitory constant Ki values were in 362.56 uM to 4.50 mM.

Table-V: Cathepsin d docking with Pepstatins
Calculation of Binding Energy, Inhibitory constant Pepstatins.

No	Ligand Name	Estimated Binding Energy Kcal/mol	Estimated Inhibition Constant Ki.	H-bonds
1.	Pepstatin	-14.70	1.26 nM	14
2.	Pepstatin analogue -1	-7.27	4.71 uM	12
3.	Pepstatin analogue -2	-8.19	1.00 uM	15
4.	Pepstatin analogue -3	-6.67	12.99 uM	9
5.	Pepstatin analogue -4	-5.20	153.45 uM	21
6.	Pepstatin analogue -5	-8.82	343.62 nM	4

The predicted binding energy of all the Pepstatins were in between - 5.20 Kcal/mol to -10.70 Kcal/mol and predicted inhibitory constant Ki values were in between 1.26 nM to 153.45 uM.

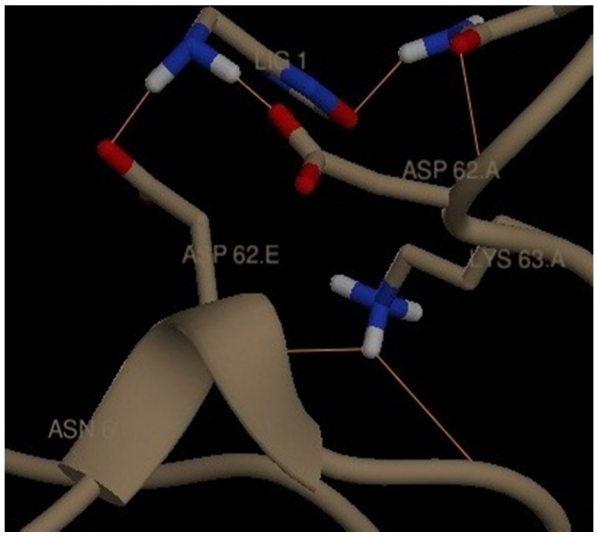


Fig: 1**:** 3D Interaction of 1,2,4-Oxadiazole-3-methylamine with cathepsin d active site residues, coloured straight lines representing the hydrogen bonds.

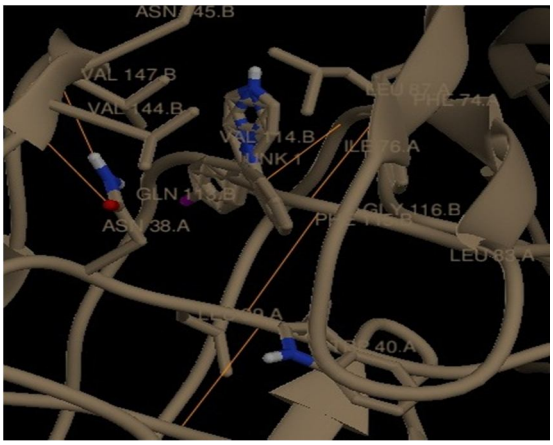


Fig**:**2 -3D Interaction of Norchlorcyclizine with cathepsin d active site residues, coloured straight lines representing the hydrogen bonds formation.

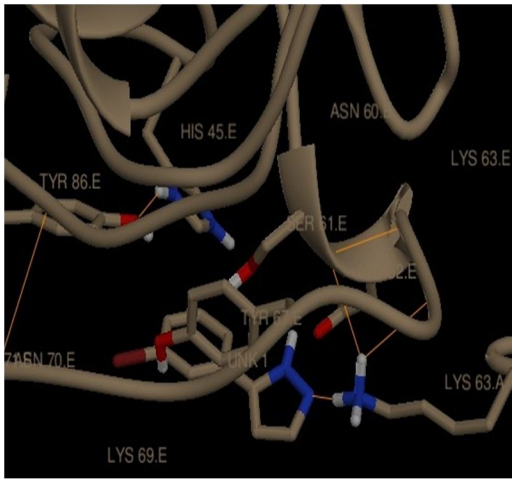


Fig**:** 3- 3D Interaction of 3-(3-Bromophenyl)-1H-pyrazole with cathepsin d active site residues, coloured straight lines representing the hydrogen bonds formation.

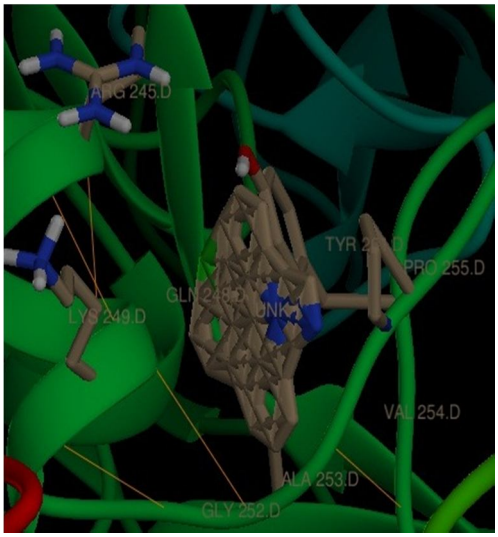


Fig**:** 4 - 3D Interaction of clotrimazole with cathepsin d active site residues, coloured straight lines representing the hydrogen bonds formation

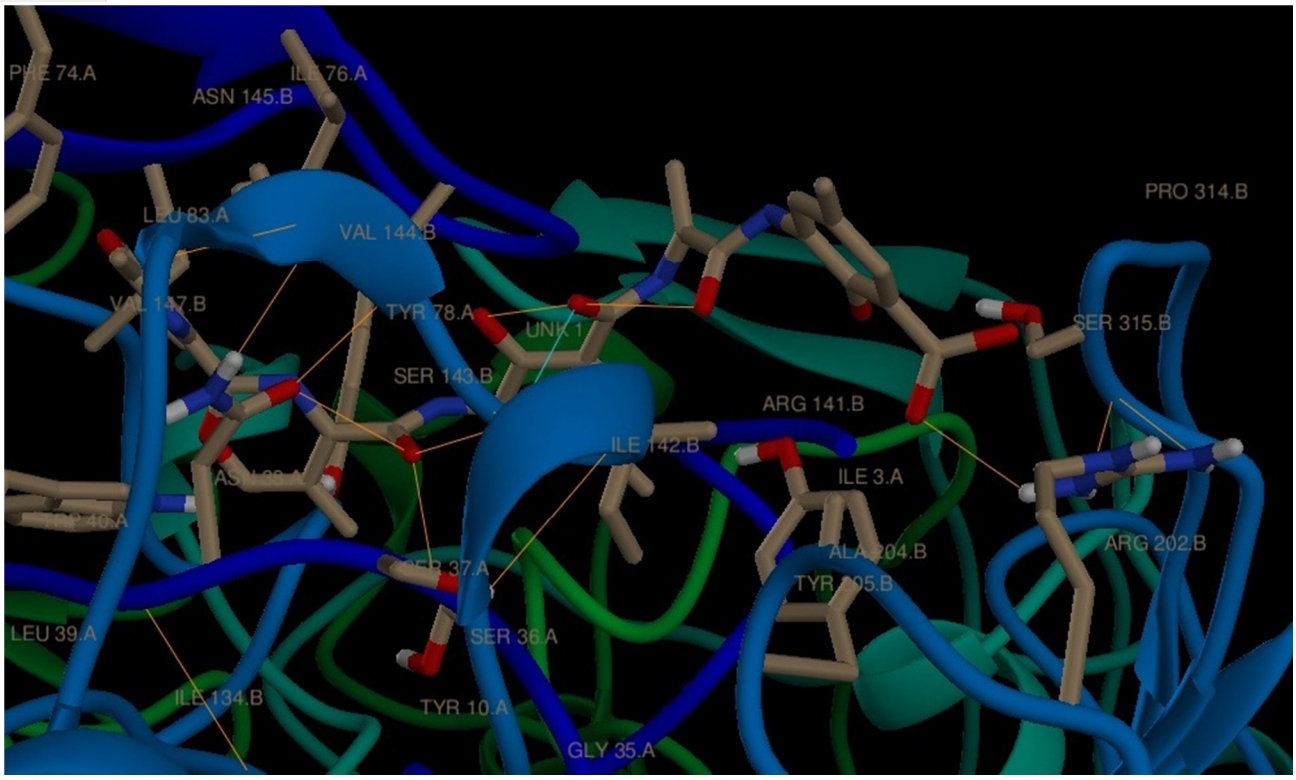


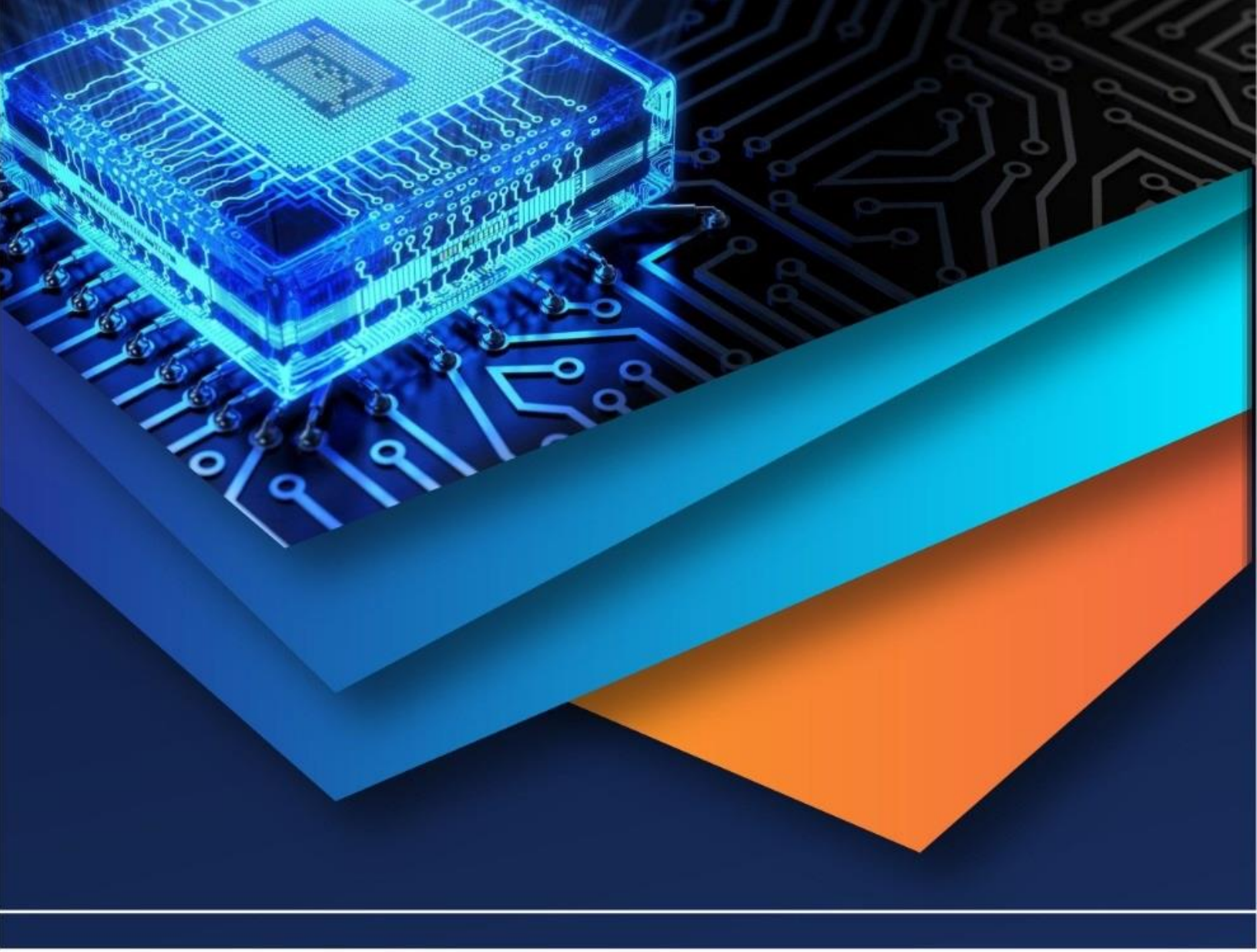
Fig**:** 5 - 3D Interaction of Pepstatin with cathepsin d active site residues, coloured straight lines representing the hydrogen bonds formation

IV. CONCLUSION

Apart from Pepstatin, Norchlorcyclizine was observed to be inhibiting cathepsin d in nanomolar range. and few amino acids in cathepsin d are actively interacting with inhibitor molecules are VAL 114, VAL 147, VAL 144, SER 143, ILE 134, TYR 10, SER 36. These docking studies helps in the exploration of cathepsin d active site aminoacids which takes part in the binding and interaction with the inhibitor molecules. By studying wide range of compounds as cathepsin d inhibitors, helps us in the development of new therapeutic agents by focusing on new synthetic approaches.

REFERENCES

[1] Kornfeld S. and Meilman I. 1989. The biogenesis of lysosomes. Annu. Rev. Cell Biol., 5:483-525.
[2] Peters P. J, Neeijes J. J, Oorschot V, Ploegh H. L, Geuze H. J, 1991 Segregation of MHC class II molecules from MHC class I molecules in the Golgi complex for transport to lysosomal compartments. Nature. 349: 669-676.
[3] Gopalan P, Dufrense M. J, Warner A. H, 1987. Thiol protease and Cathepsin D activities in selected tissues and cultured cells from normal and dystrophic mice.Can. J. Physiol. Pharmacol. 65:124-129
[4] Cataldo A. M and Nixon R. A,1990. Enzymatically active lysosomal proteases are associated with amyloid deposits in Alzheimer brain. Proc. Natl. Acad. Sci. USA 87, 3861-3865.
[5] Kenessey A, Banay-Schwartz M, DeGuzman T, Lajtha A. J, 1989.Increase in cathepsin D activity in rat brain in aging. Neurosci. Res. 23:454-456.
[6] Davies, D. R. (1990) The structure and function of the aspartic proteinases, Annual review of biophysics and biophysical chemistry 19, 189-215.
[7] Dunn, B. M., Scarborough, P. E., Lowther, W. T., and Rao-Naik, C. (1995) Comparison of the active site specificity of the aspartic proteinases based on a systematic series of peptide substrates, Advances in experimental medicine and biology 362, 1-9.



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