

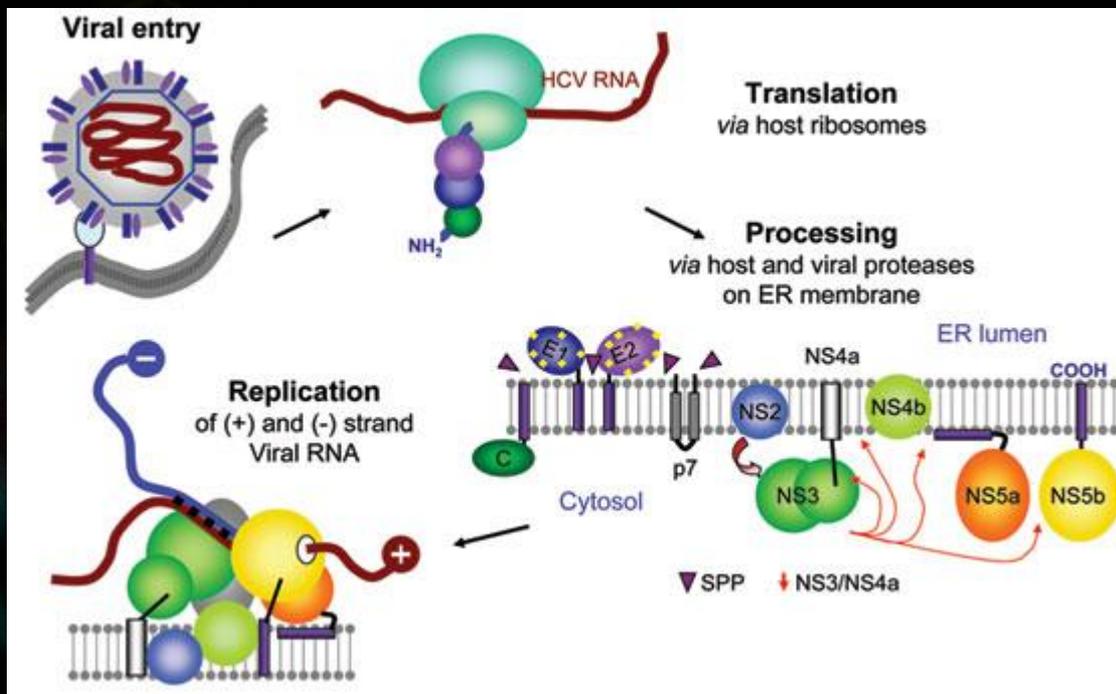
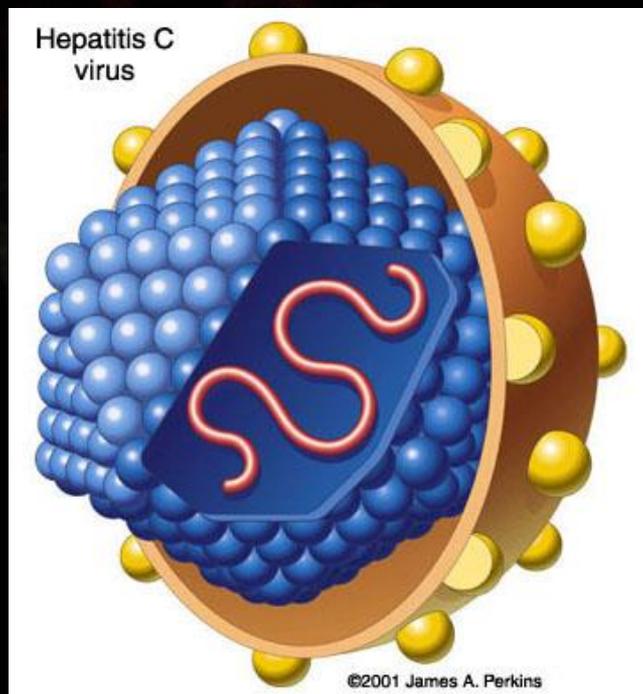


HCV RNA polimerasi (NS5B):
relazioni struttura-attività tridimensionali
di tipo quantitativo (3-D QSARs) e
valutazione della capacità predittiva.

Relatore: Dr. Rino Ragno

Laureanda: Teresa Fantasia

HCV

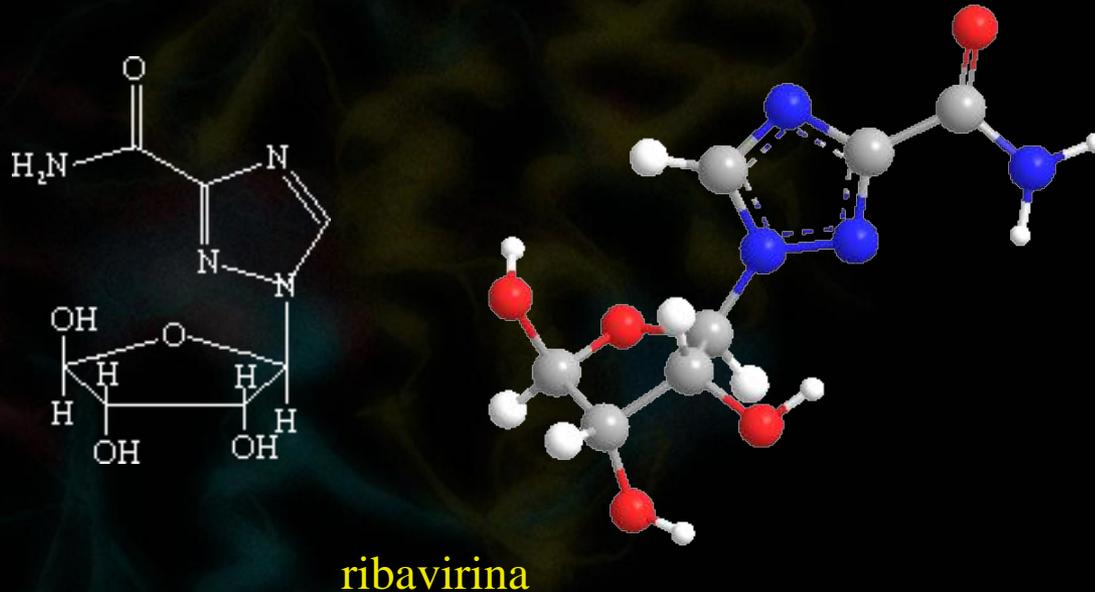


STANSFIELD *ET AL.*, BIOORGANIC & MEDICINAL CHEMISTRY LETTERS, 2007.

MANNS *ET AL.*, NATURE REVIEWS, 2007.

HCV - Terapia attuale

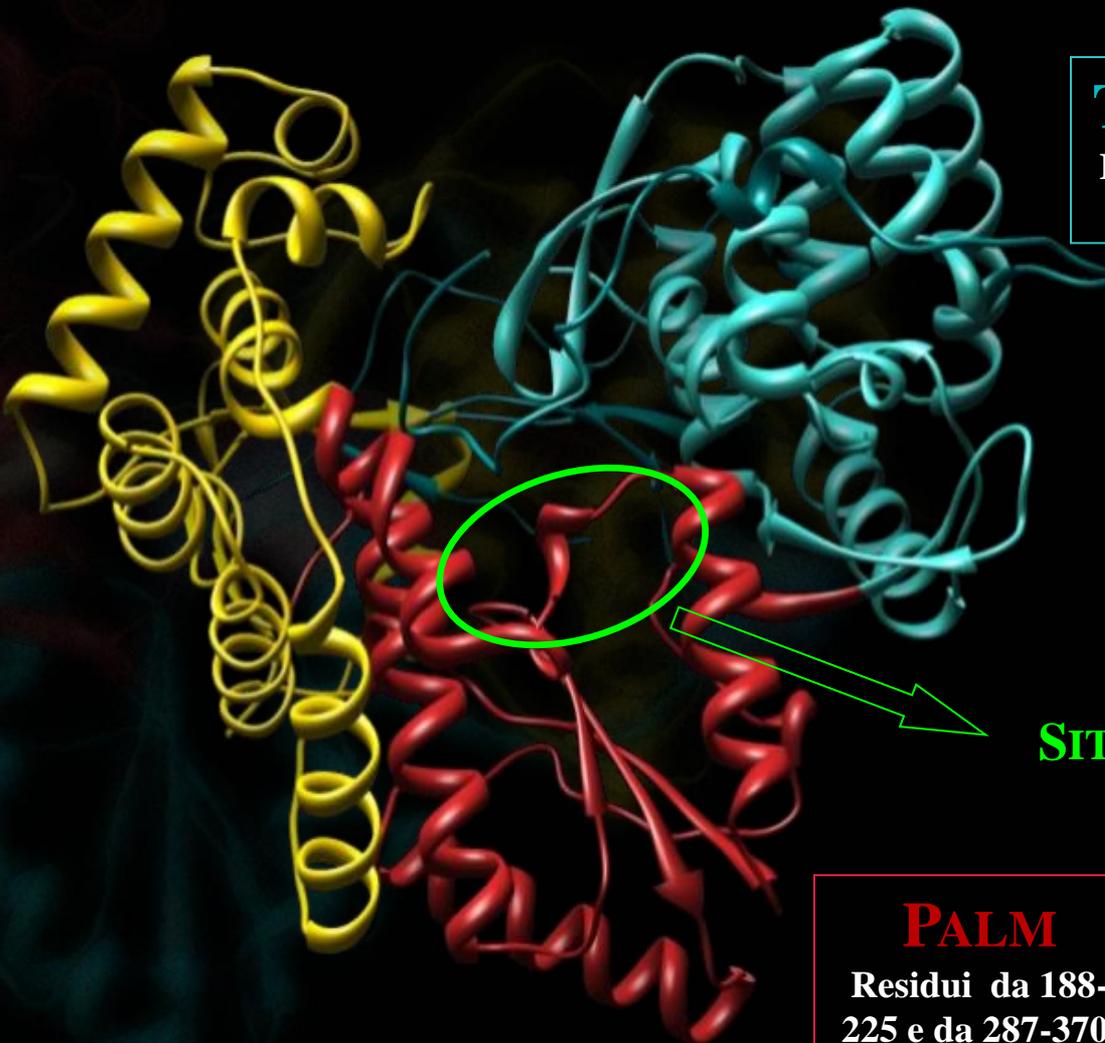
- Monoterapia con IFN- α
- Associazione IFN- α /ribavirina



**NECESSITÀ DI NUOVI INIBITORI POTENTI E AD AMPIO
SPETTRO D'AZIONE.**

NS5B (RdRp)

FINGERS
Residui da
1 - 187



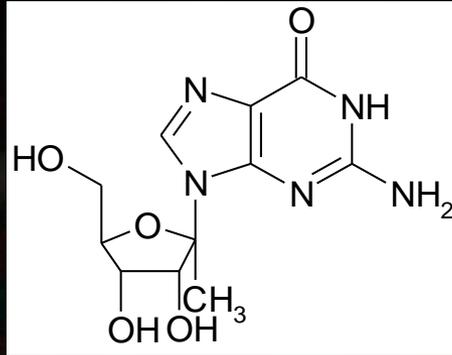
THUMB
Residui da
371 a 528.

PALM
Residui da 188-
225 e da 287-370.

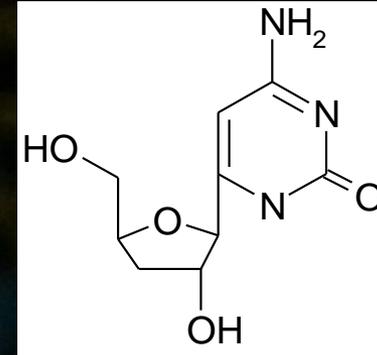
SITO ATTIVO

Inibitori nucleosidici (NI)

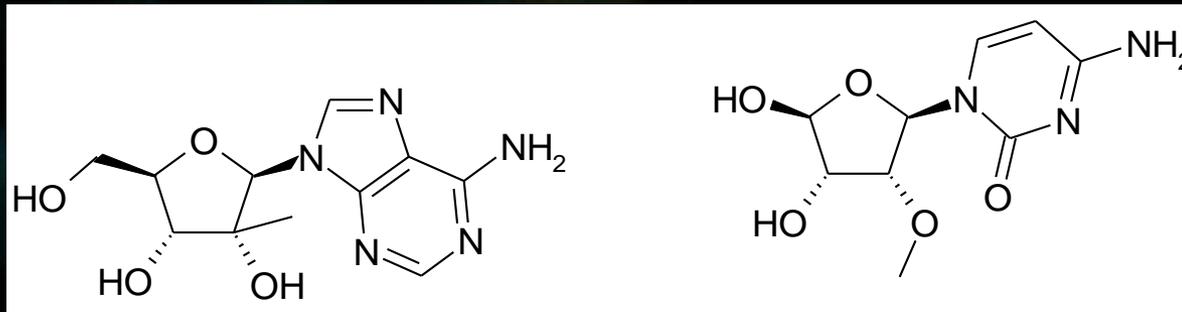
- AGISCONO SUL SITO ATTIVO IN MODO COMPETITIVO
- CONVERSIONE IN TRIFOSFATO
- INCORPORAZIONE NELLA CATENA DELL'ACIDO NUCLEICO → PREMATURA TERMINAZIONE SINTESI RNA VIRALE



β -D-2'-metilribofuranosilguanosina

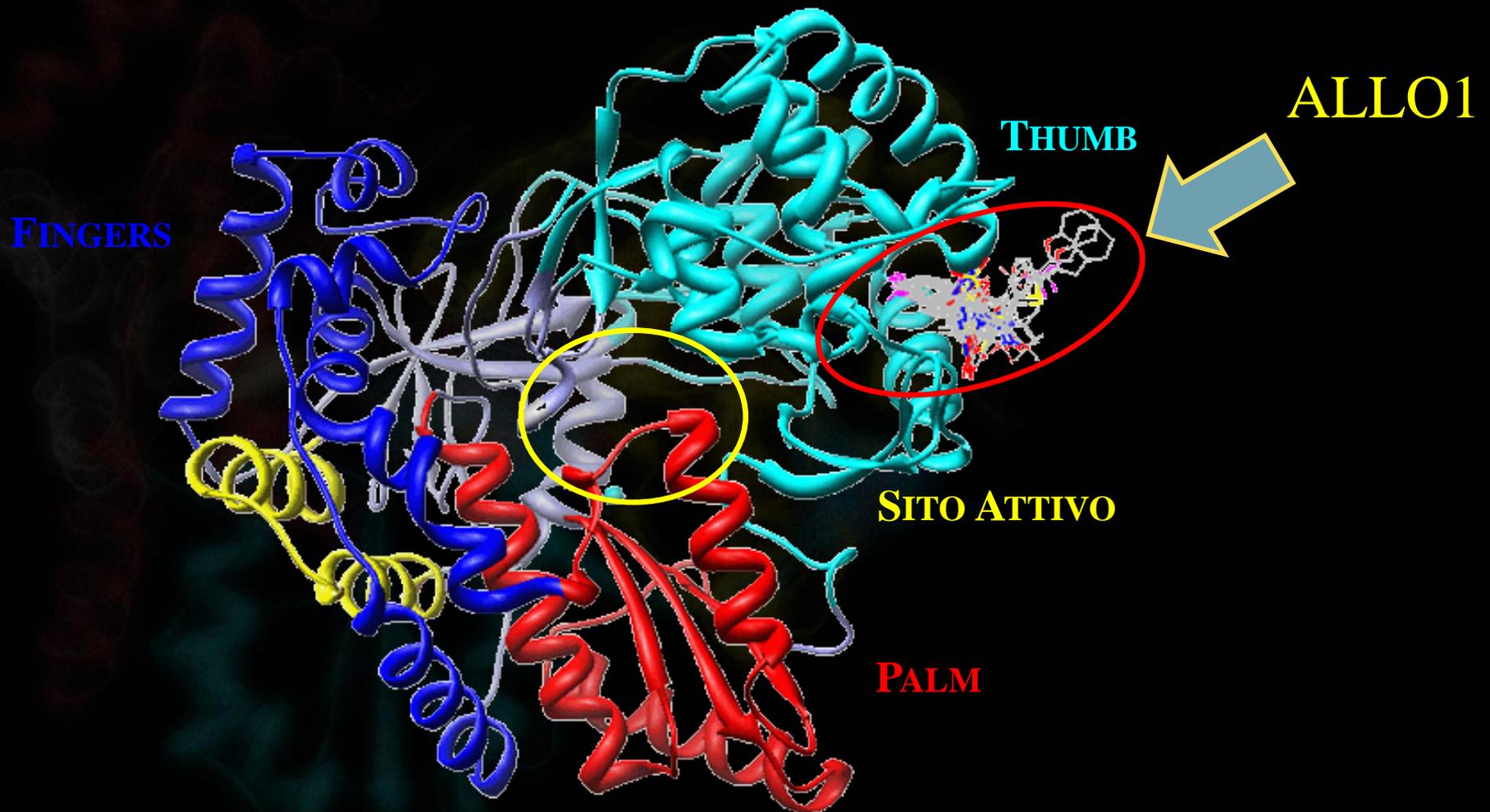


3'-deossinucleosidi

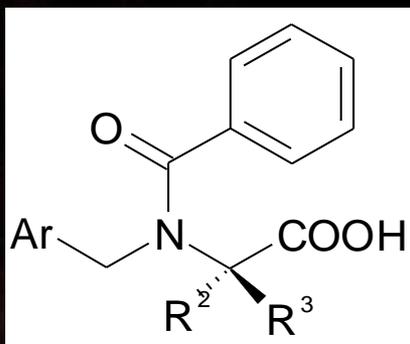


2'-C-metiladenosina e 2'-O-metilcitidina

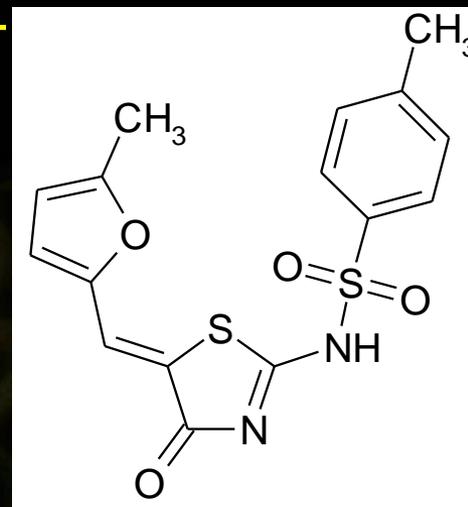
Inibitori allosterici del thumb (1)



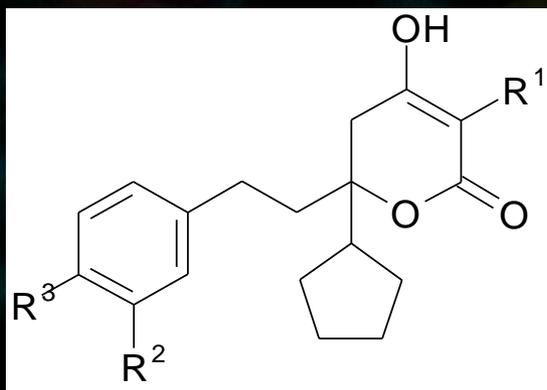
Inibitori allosterici del thumb (1)



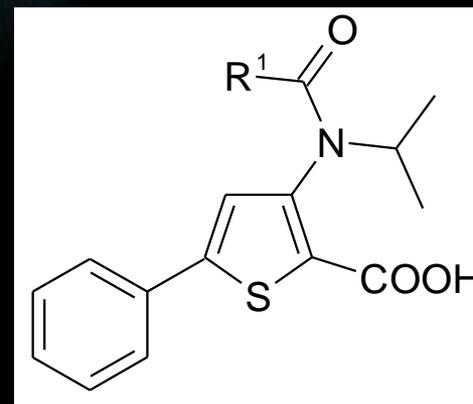
FENILALANINA N,N-DISOSTITUITA



TIAZOLON-DERIVATI

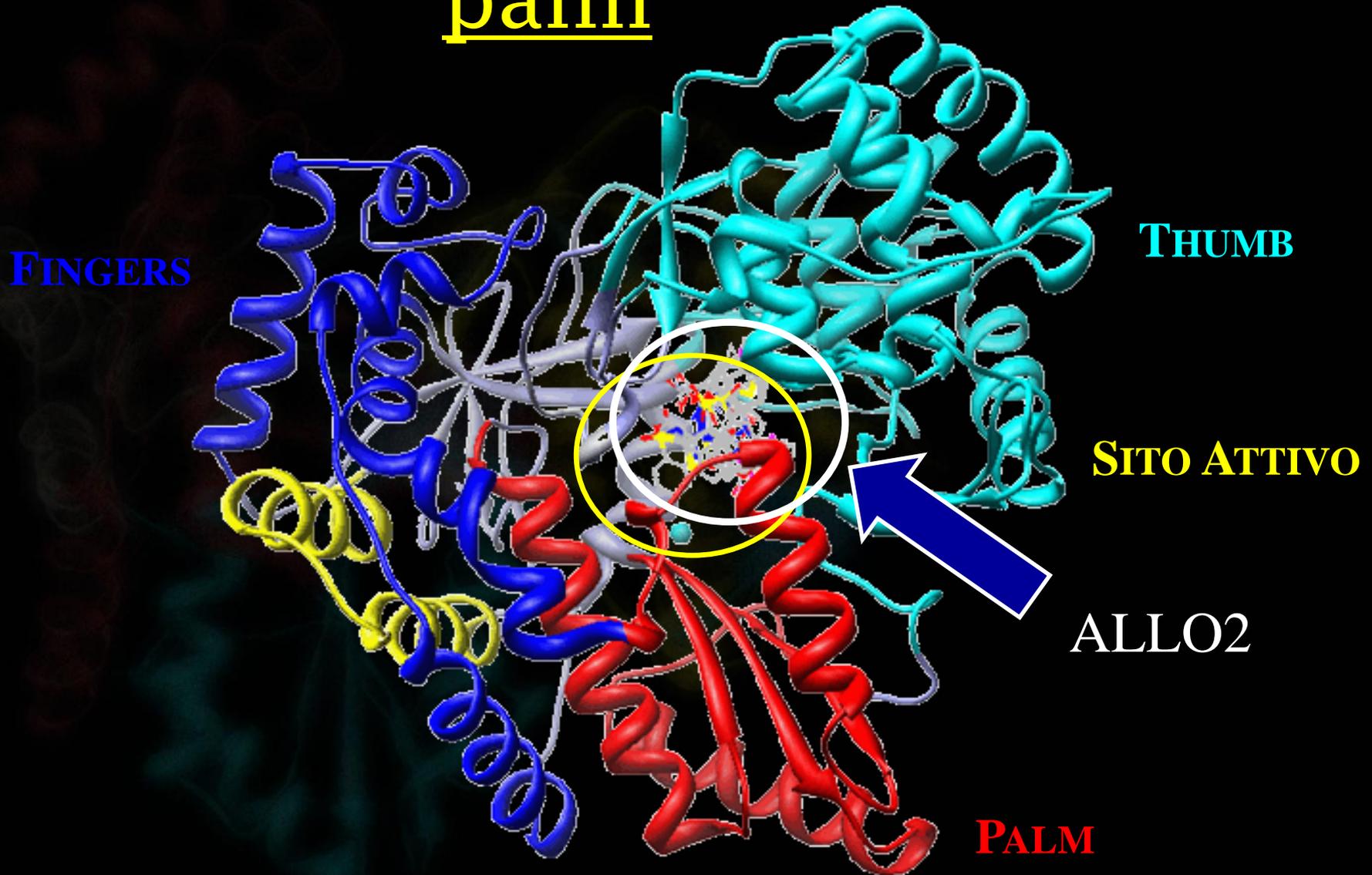


DIIDROPIRONI

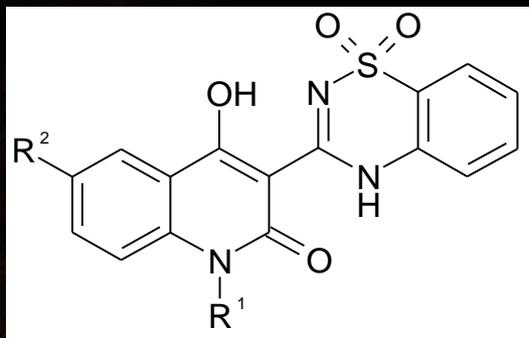


ACIDI TIOFEN-2-CARBOSSILICI

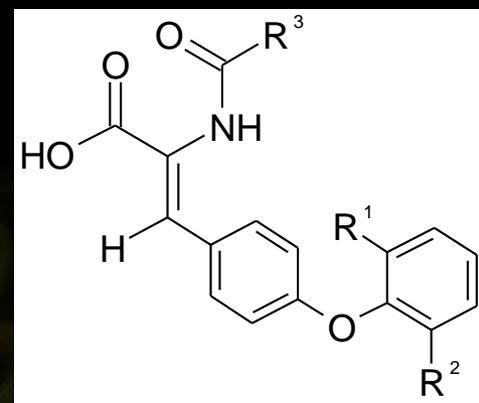
Inibitori allosterici del palm



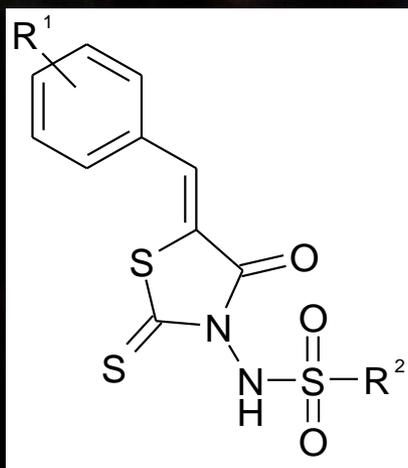
Inibitori allosterici del palm



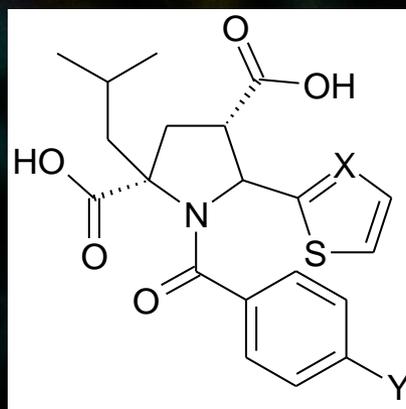
BENZOTIADIAZIN-CHINOLINONI



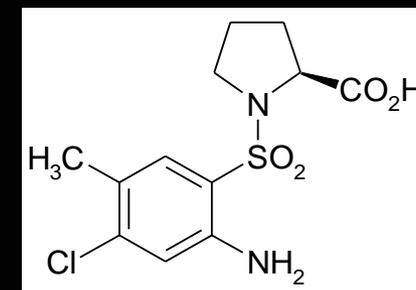
DERIVATI DELL'ACIDO ACRILICO



BENZILIDENI

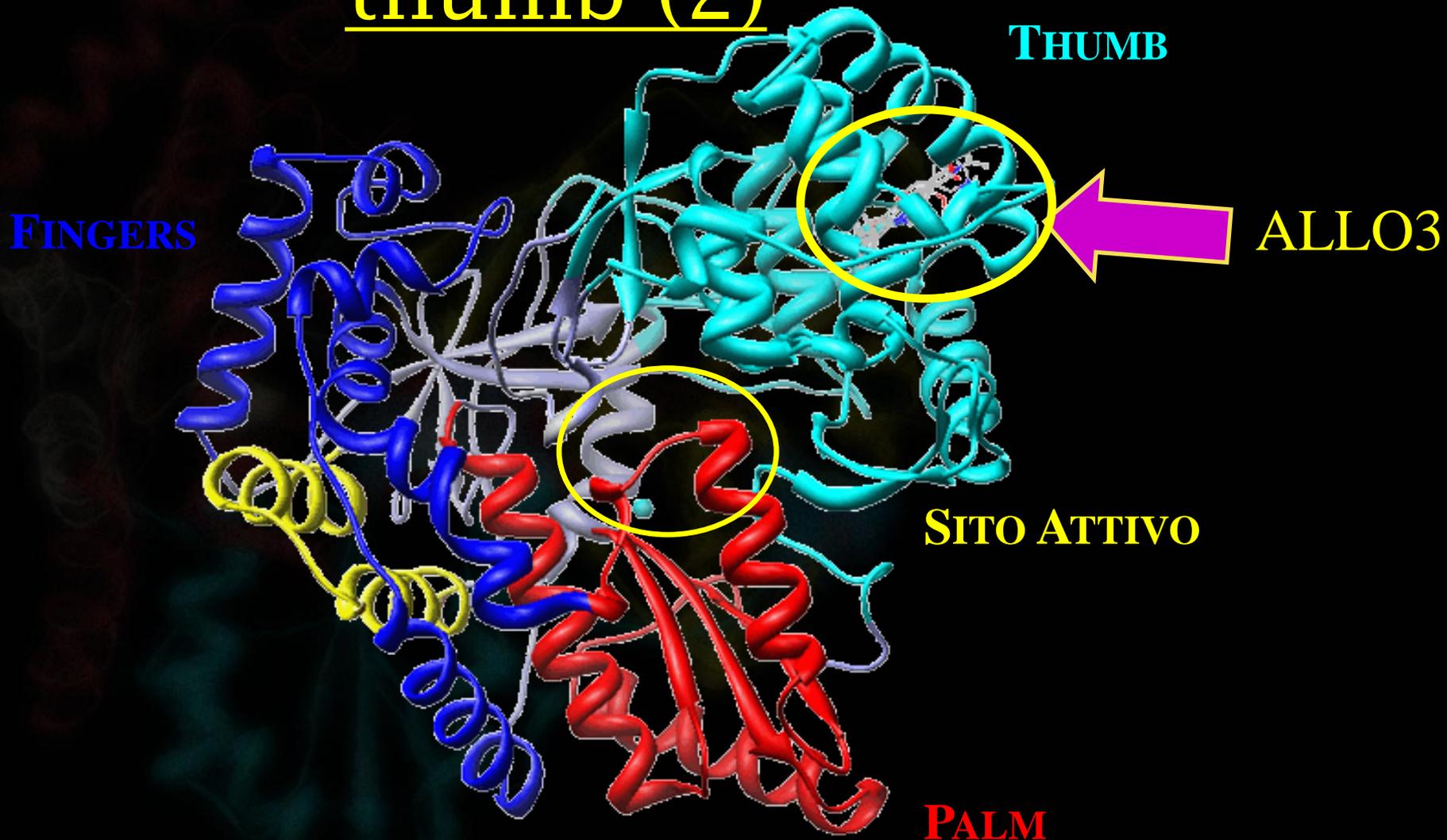


ACIL-PIRROLIDINE

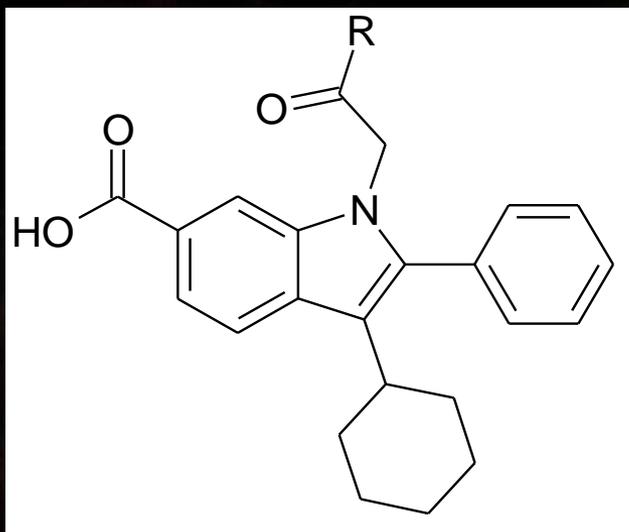


PROLINA SOLFONAMIDI

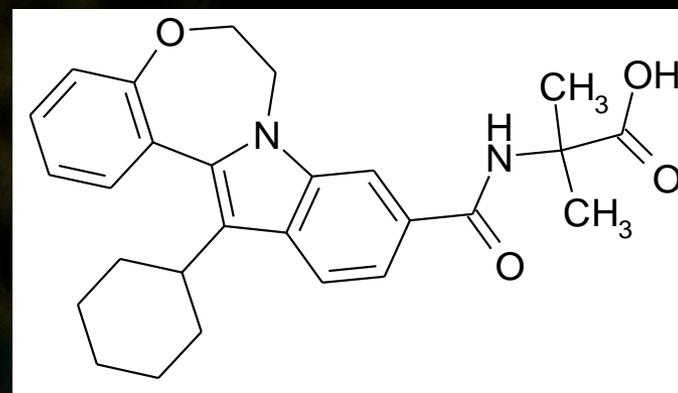
Inibitori allosterici del thumb (2)



Inibitori allosterici del thumb (2)



INDOLI N-SOSTITUITI



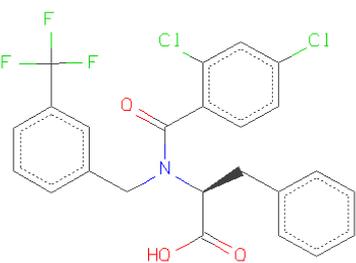
COMPOSTI TETRACICLICI

Costruzione dei modelli 3-D QSAR

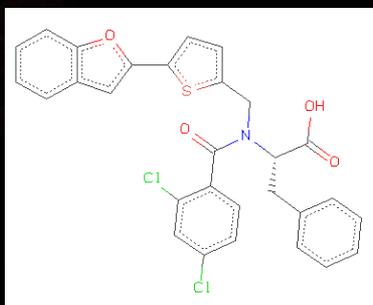


- SCELTA DEL TRAINING SET
- COSTRUZIONE DEI MODELLI MOLECOLARI
- ALLINEAMENTO DELLE MOLECOLE
- CALCOLO DEI CAMPI DI INTERAZIONE MOLECOLARE (MIF)
- ELABORAZIONE STATISTICA DEI MODELLI 3-D QSAR (PLS/GOLPE)
- VALIDAZIONE DEI MODELLI 3-D QSAR
- INTERPRETAZIONE DEI RISULTATI

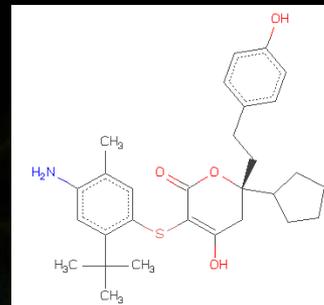
PRIMO TRAINING SET: 15 LIGANDI CO-CRISTALLIZZATI NEL SITO ALLOSTERICO SITUATO NEL DOMINIO THUMB (ALLO1).



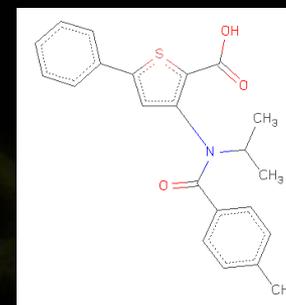
1NHU



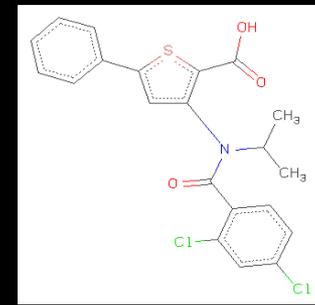
1NHV



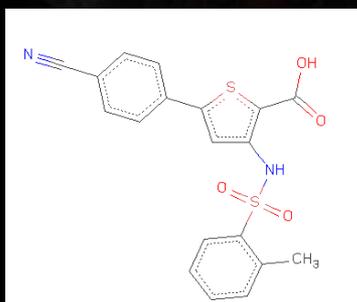
1OS5



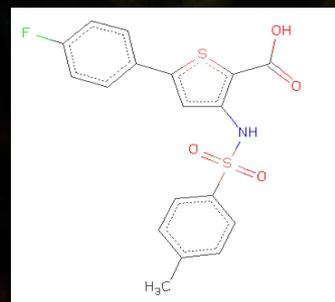
1YVX



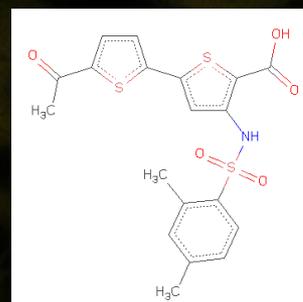
1YVZ



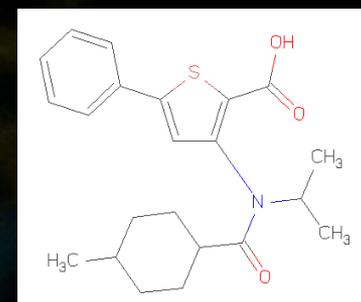
2D3U



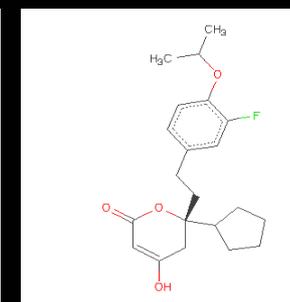
2D3Z



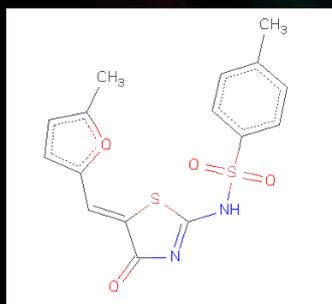
2D41



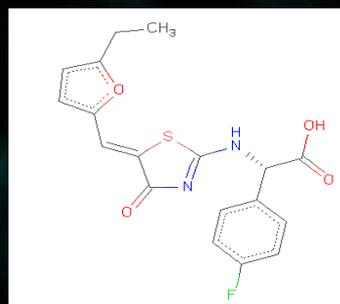
2GIR



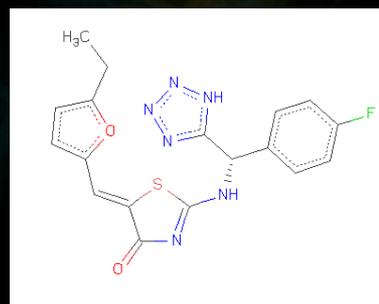
2HAI



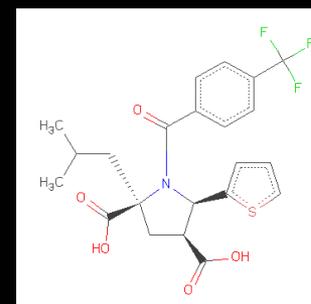
2HWH



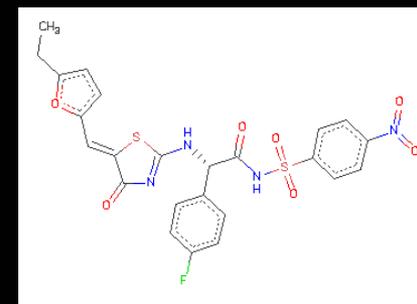
2HWI



2I1R

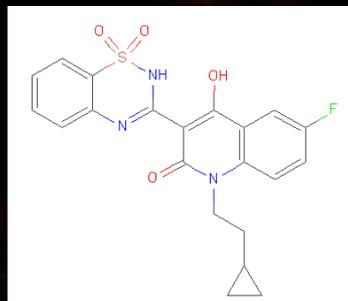


2JCO

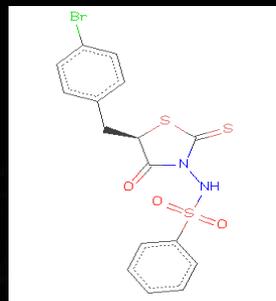


2O5D¹³

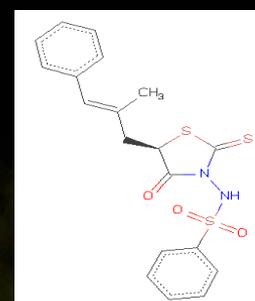
SECONDO TRAINING SET: 10 LIGANDI CO-CRISTALLIZATI NEL SITO ALLOSTERICO SITUATO NEL DOMINIO PALM (ALLO2).



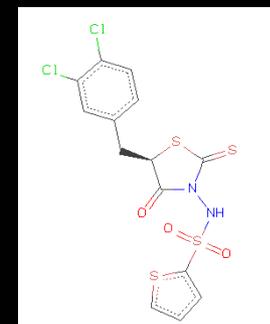
2GIQ



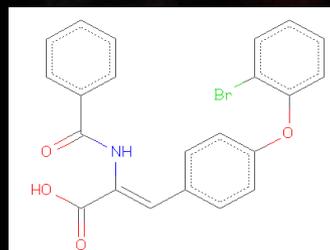
2AWZ



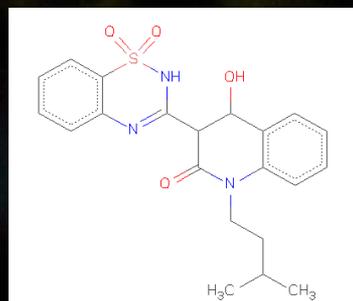
2AXO



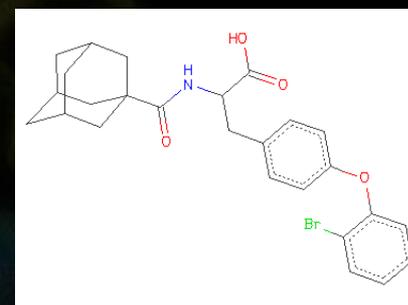
2AX1



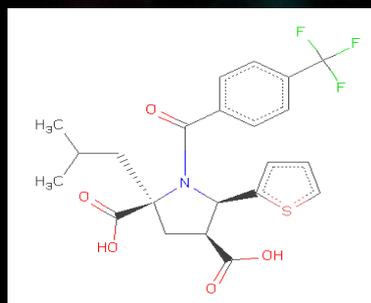
1YVF



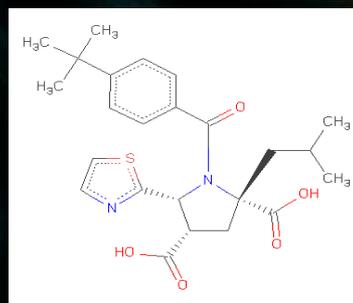
2FVC



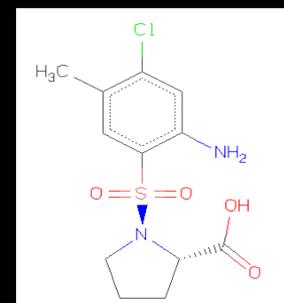
1Z4U



2JC0



2JC1



2GC8

Costruzione dei modelli molecolari

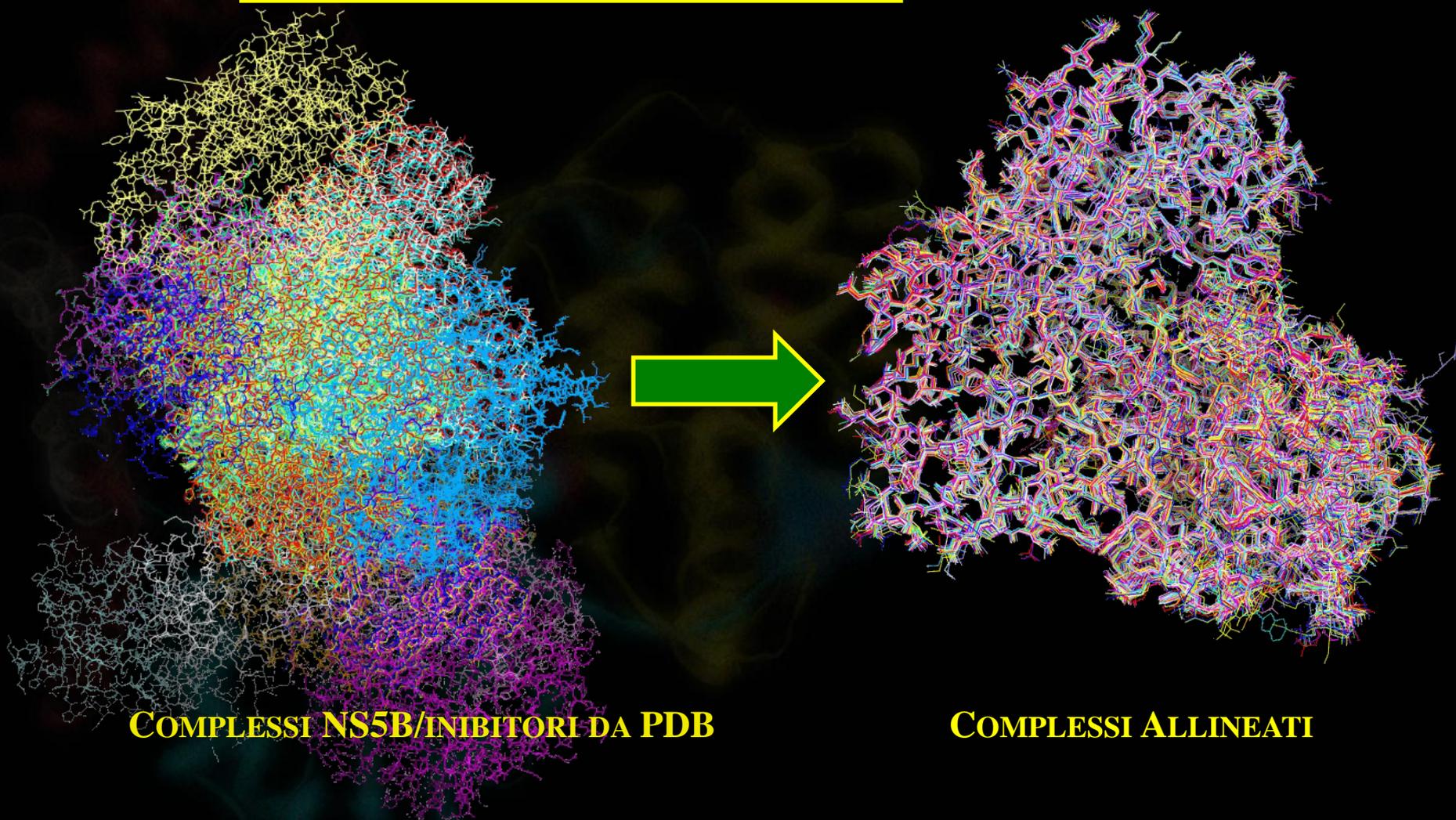


MINIMIZZAZIONE (AMBER9)

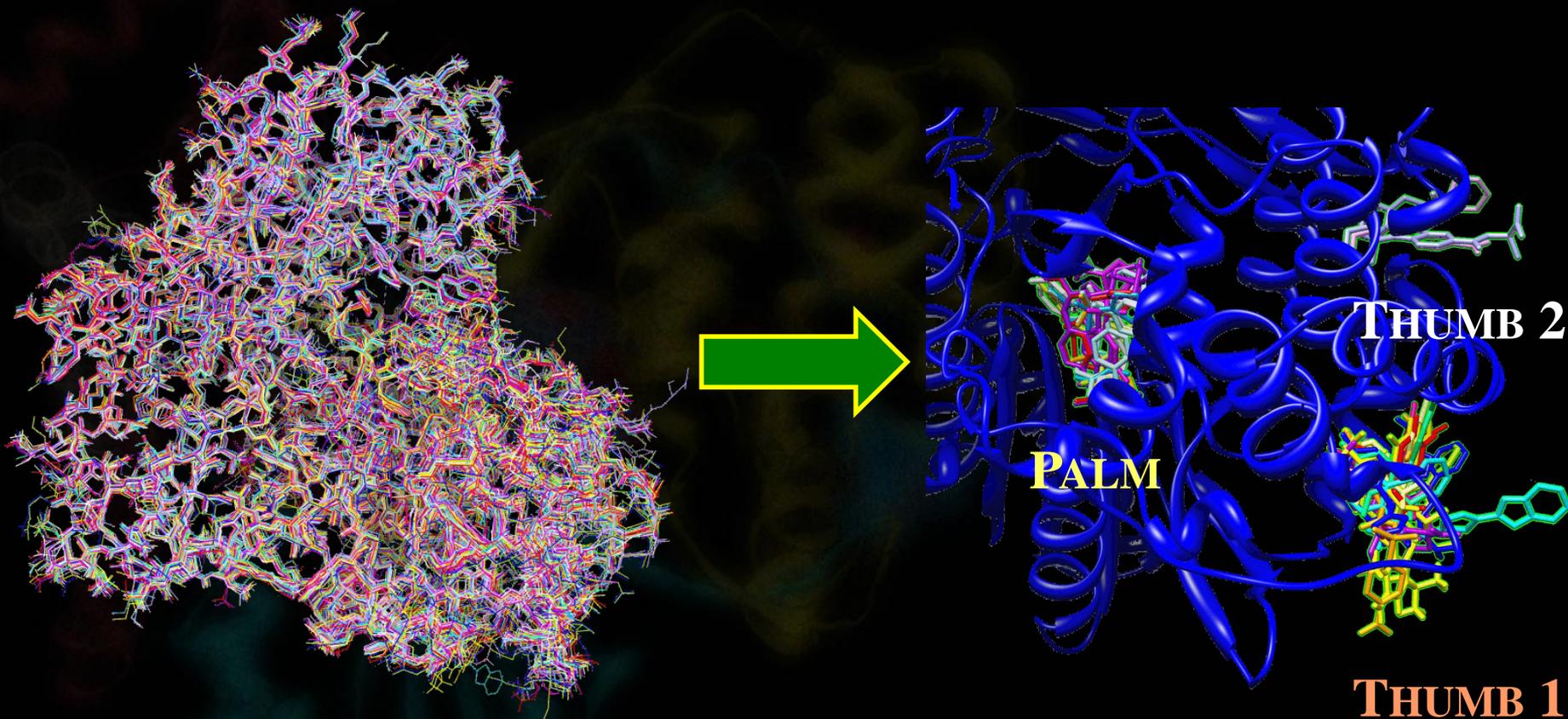


COMPLESSI OTTIMIZZATI GEOMETRICAMENTE
E STRUTTURALMENTE

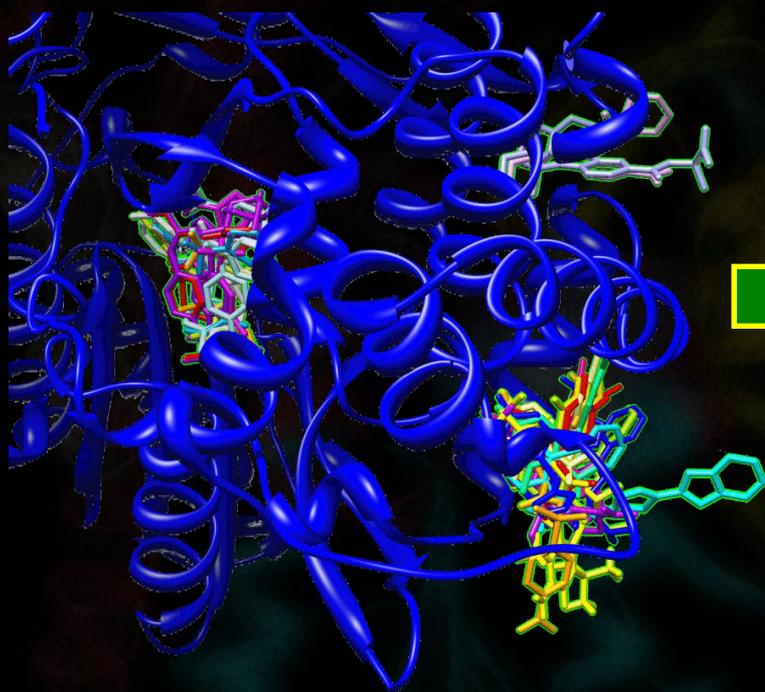
Allineamento Structure - based



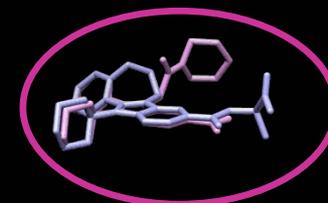
Alineamento Sperimentale



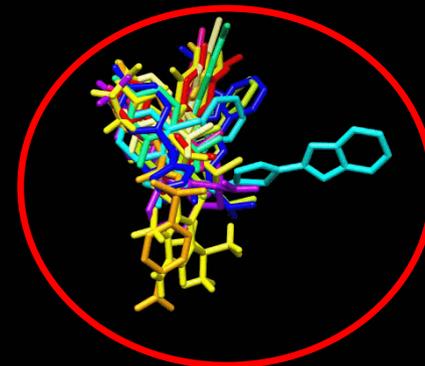
Allineamento Sperimentale



PALM



THUMB 2



THUMB 1

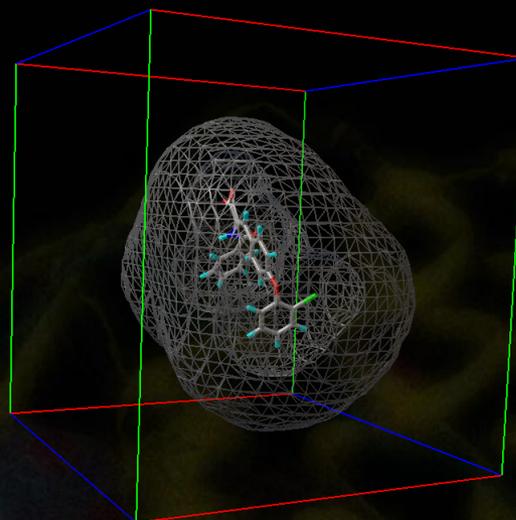
Allineamento Sperimentale



**PALM
(ALLO2)**



**THUMB 1
(ALLO1)**



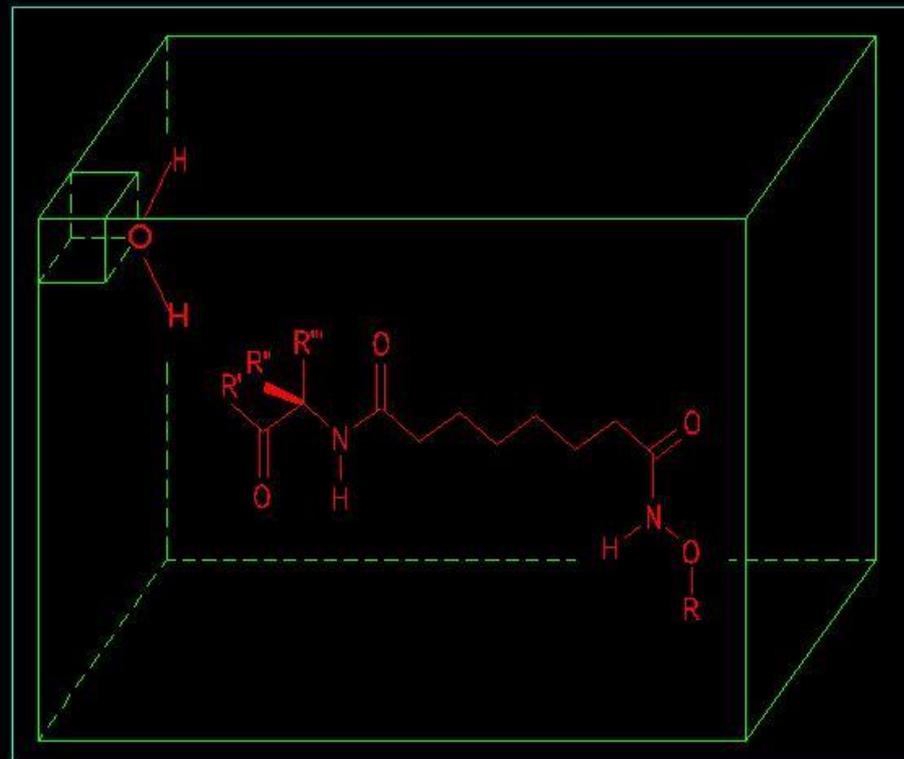
GRID/GOLPE

Modelli 3-D QSAR

Calcolo dei campi d'interazione molecolare (MIF)

GRID

Interazioni energetiche
probe-target per ogni
punto della griglia
costruita attorno al
target



Elaborazione statistica dei modelli 3-D QSAR



PLS/GOLPE

I dati ottenuti da GRID vengono importati su
GOLPE → relazione x/y

Selezione delle variabili → eliminate
informazioni poco significative o ripetitive

Risultati Statistici dei modelli

3-D QSAR

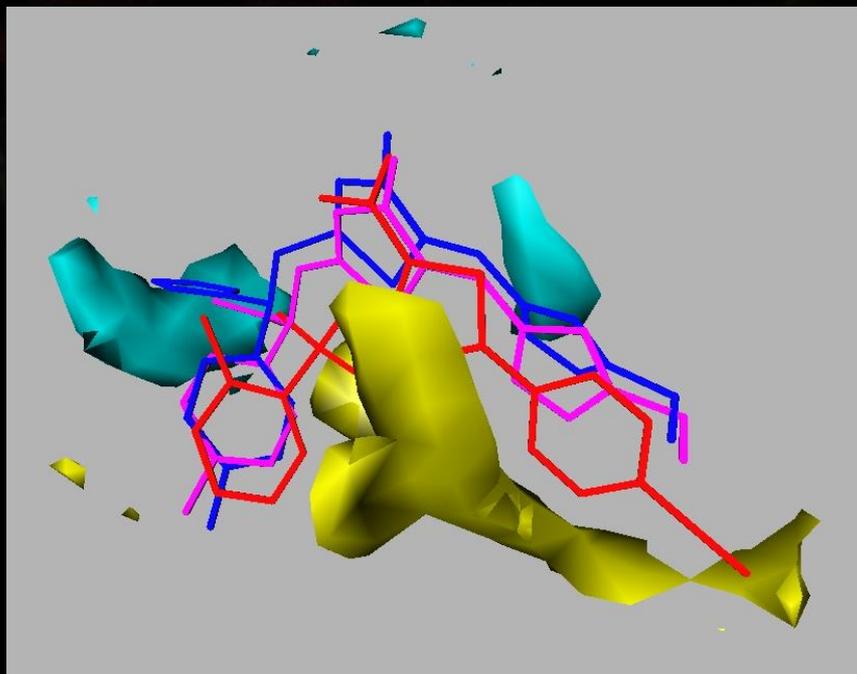


ALLO1	C1=	ALLO2	C3
r^2	0.99	r^2	0.98
q^2	0.68	q^2	0.50
SDEP _{cv}	0.32	SDEP _{cv}	0.68
PC	3	PC	2

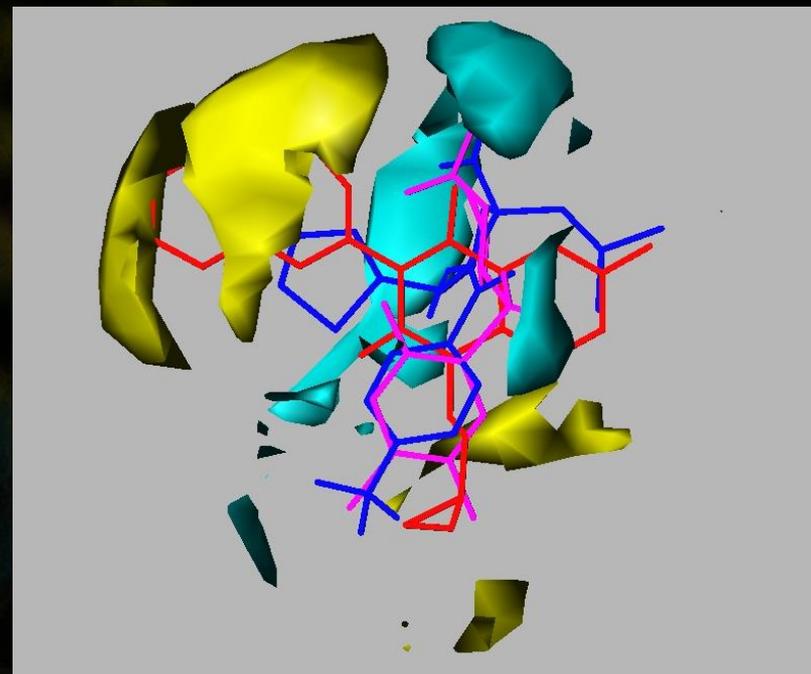
selezione del 10

ALLO

Mappe di correlazione

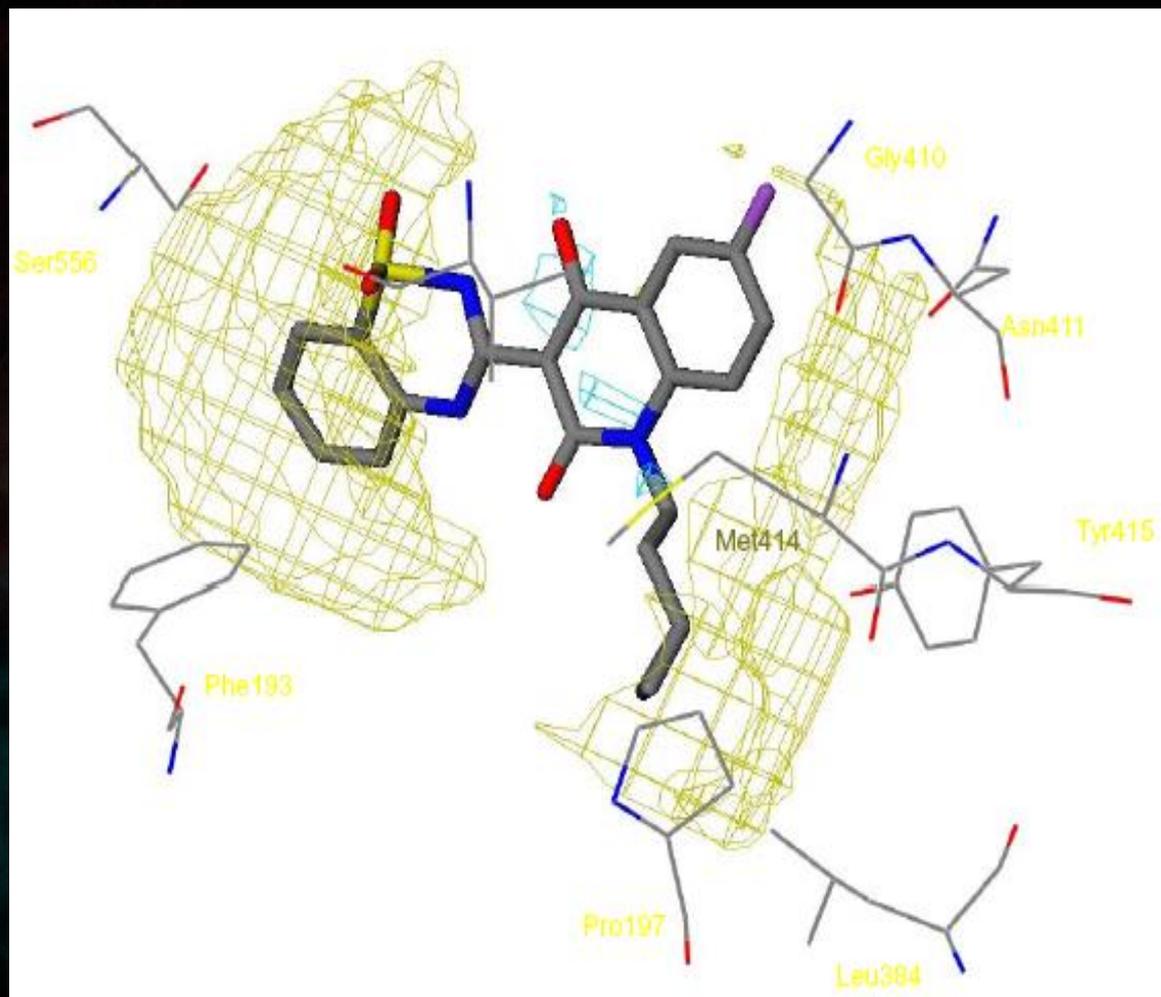


PLS-coefficients ottenuto dal modello ALLO1
utilizzando il probe C1=



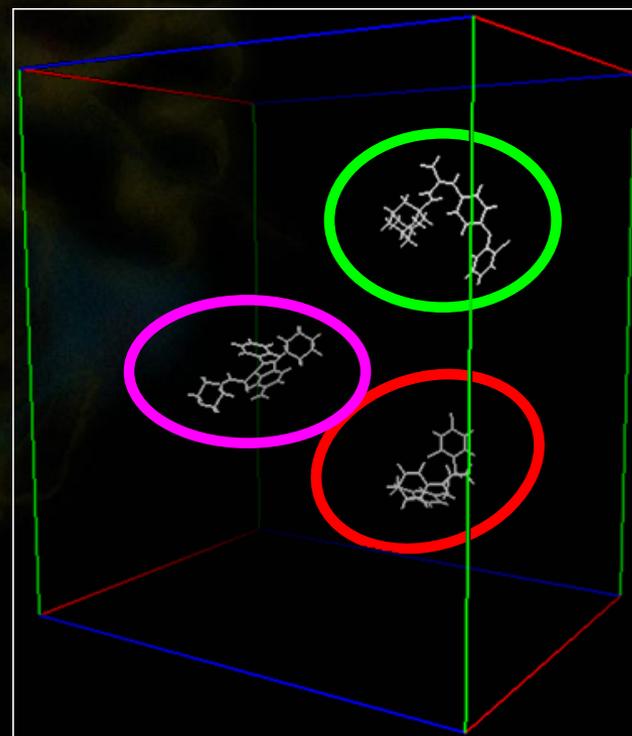
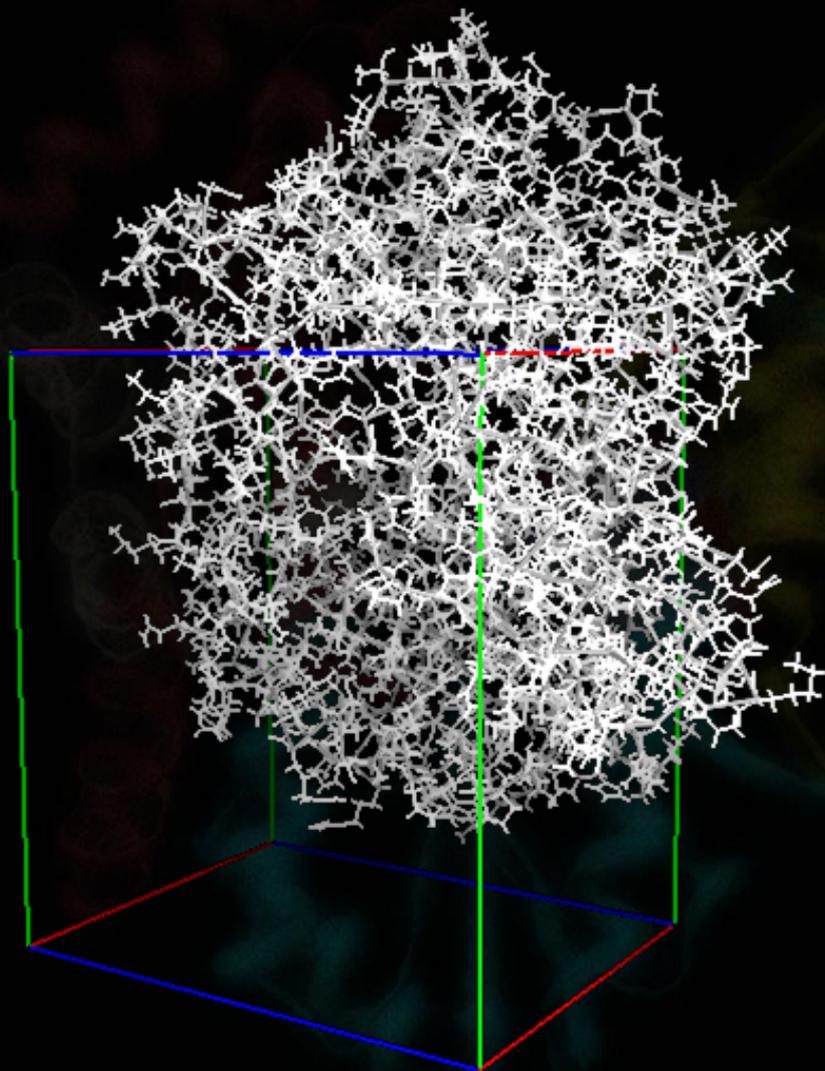
PLS-coefficients ottenuto dal modello ALLO2
utilizzando il probe C3

GRID - GOLPE ACTIVITY CONTRIBUTIONS PLOT



ACTIVITY CONTRIBUTION PLOT

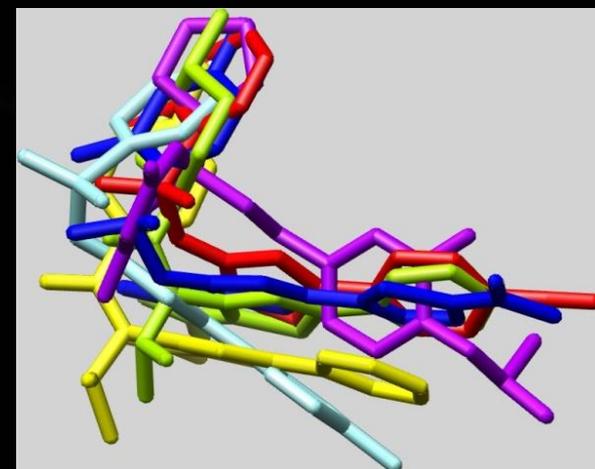
Docking (Autodock 4.0.1)



Validazione della capacità predittiva

TEST SET ESTERNI { ALLO1 (81 molecole)
ALLO2 (223 molecole)

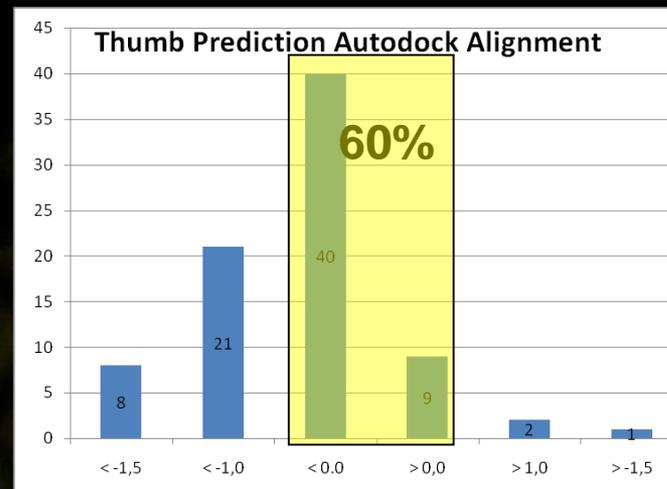
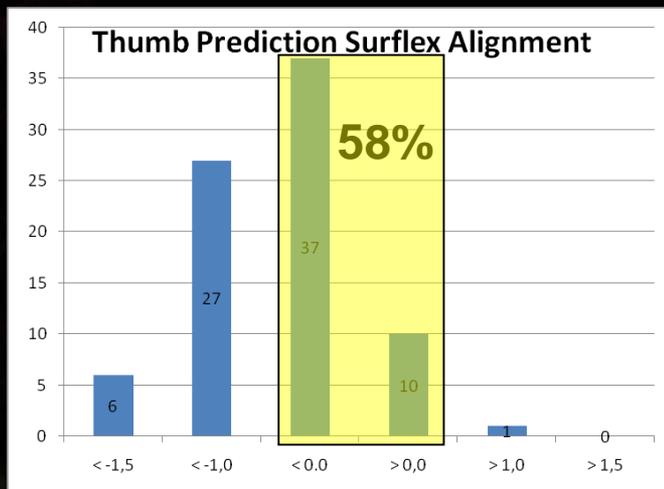
Allineamento (SURFLEX) → maxs



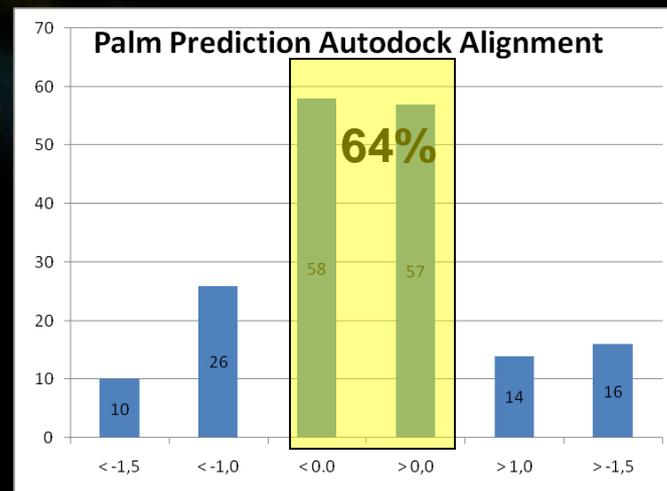
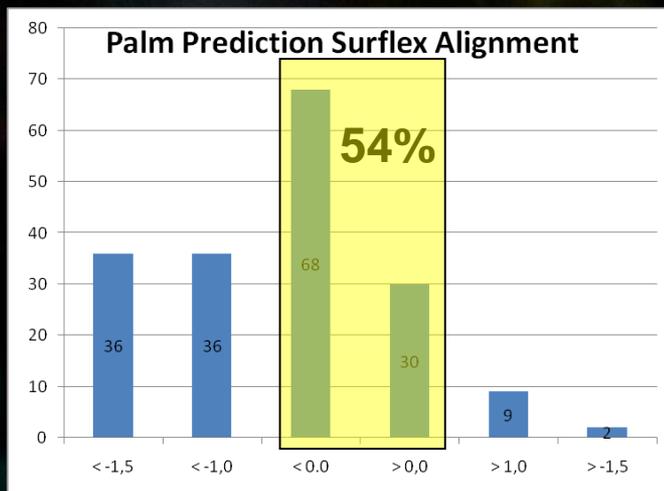
Cross-docking (AUTODOCK 4.0.1.) → Best Cluster

Predittività dei modelli 3-D QSAR

Thumb Test Set ALLO1

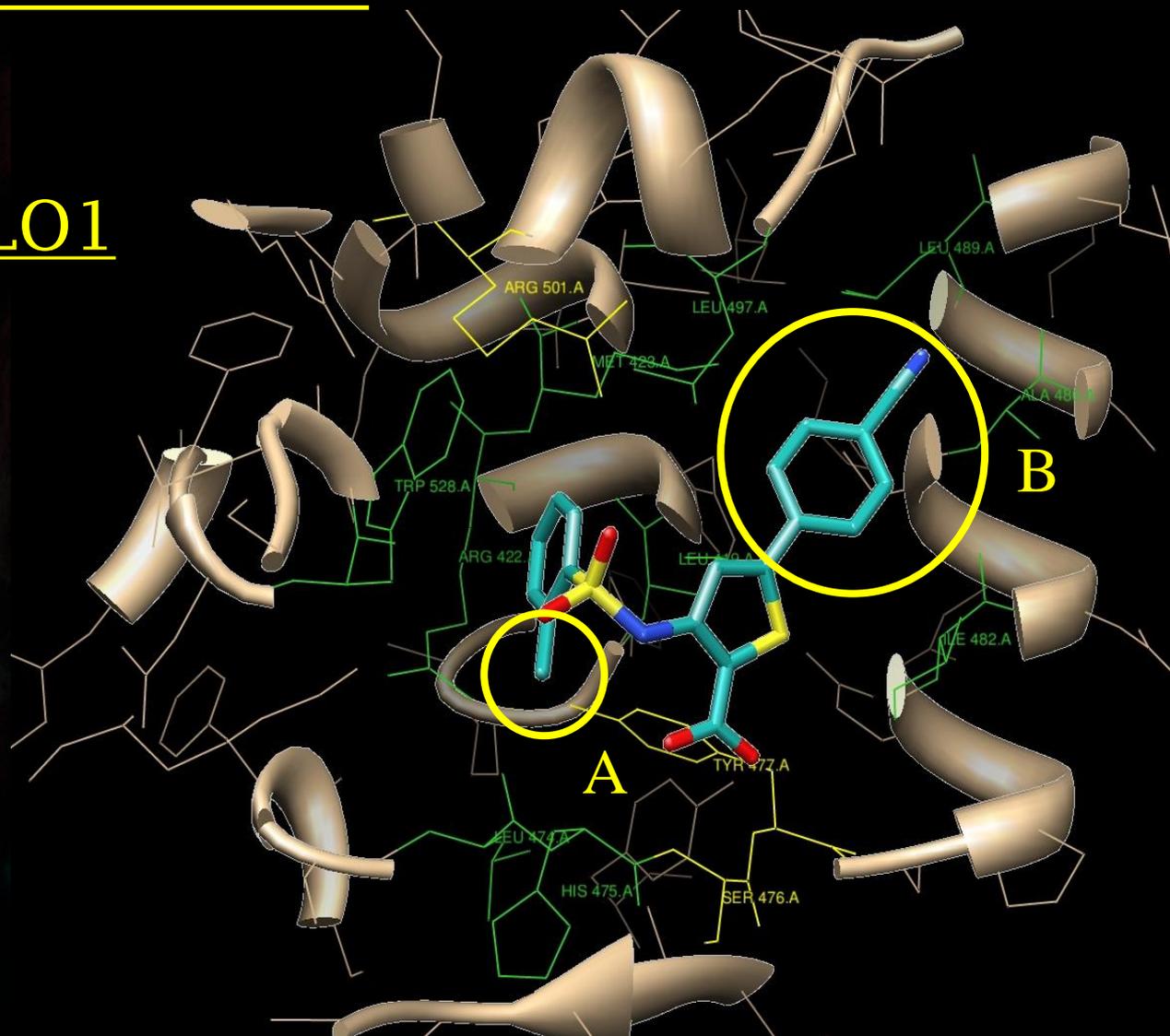


Palm Test Set ALLO2



Conclusioni

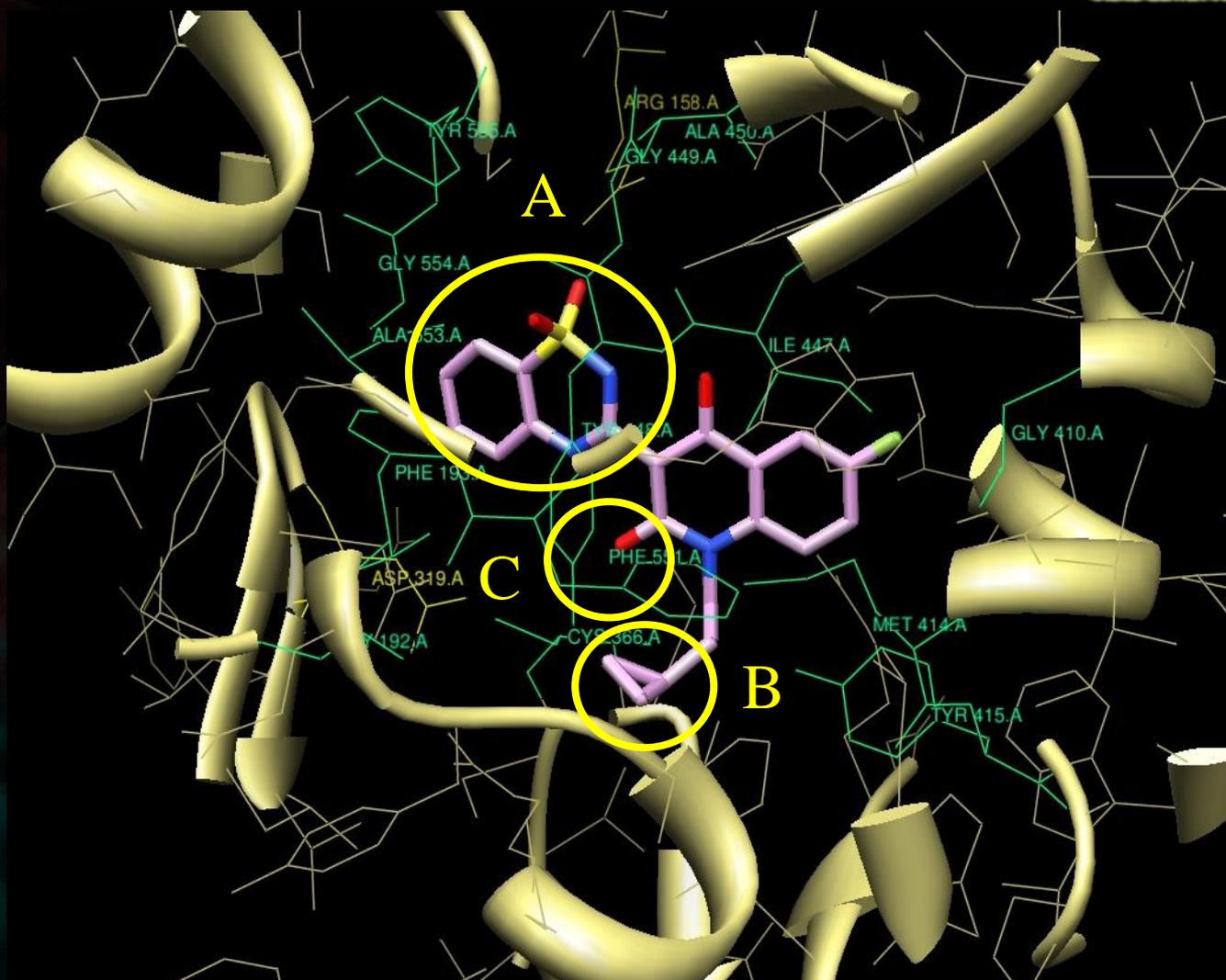
ALLO1



Molecola più attiva del modello ALLO1 in cui sono evidenziate le possibili zone di maggiore interesse biologico

Conclusioni

ALLO2



Molecola più attiva del modello ALLO2 in cui sono evidenziate le zone di maggior interesse biologico