

Technical Note

TN2007-03 (July 2007)

Reagent: Macaque-reactive anti-CD73 antibodies

Summary: Macaque monkeys have >98% amino acid sequence homology with human CD73. Rhesus (*M. mulatta*) and cynomolgus (*M. fascicularis*) differ by only 2 of 574 amino acids. All anti-human CD73 antibodies tested reacted with rhesus CD73.

Part I. CD73 sequence analysis

Method: Rhesus and cynomolgus CD73 were cloned from renal tissue and sequenced.

Alignment of rhesus, cynomolgus and human CD73 amino acid sequences

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MCPRAARAPATLLAVGALLWSAAGAWELTILHTNDVHSRLEQTSSESSKCVNASRCMGGVARLFTKVQQIRRAEPNVLLLDAGDQYQGTIWFTVYKGAE
  10      20      30      40      50      60      70      80      90     100
Rh_CD73L_Prot MCPRAARAPATLLAVGALLWSAAGAWELTILHTNDVHSRLEQTSSESSKCVNASRCMGGVARLFTKVQQIRRAEPNVLLLDAGDQYQGTIWFTVYKGAE 100
Cyno-CD73_Pro MCPRAARAPATLLAVGALLWSAAGAWELTILHTNDVHSRLEQTSSESSKCVNASRCMGGVARLFTKVQQIRRAEPNVLLLDAGDQYQGTIWFTVYKGAE 100
Hu_CD73-L_Pro MCPRAARAPATLLAVGALLWSAAGAWELTILHTNDVHSRLEQTSSESSKCVNASRCMGGVARLFTKVQQIRRAEPNVLLLDAGDQYQGTIWFTVYKGAE 100

VAHFMNALRYDAMALGNHEFDNGVEGLIEPLLKEAKFPILSANIKAKGPLASQISGLYLPYKVLPGVDEVVGVGIVGYSKETPFLSNPGTNLVFEDEITAL
 110     120     130     140     150     160     170     180     190     200
Rh_CD73L_Prot VAHFMNALRYDAMALGNHEFDNGVEGLIEPLLKEAKFPILSANIKAKGPLASQISGLYLPYKVLPGVDEVVGVGIVGYSKETPFLSNPGTNLVFEDEITAL 200
Cyno-CD73_Pro VAHFMNALRYDAMALGNHEFDNGVEGLIEPLLKEAKFPILSANIKAKGPLASQISGLYLPYKVLPGVDEVVGVGIVGYSKETPFLSNPGTNLVFEDEITAL 200
Hu_CD73-L_Pro VAHFMNALRYDAMALGNHEFDNGVEGLIEPLLKEAKFPILSANIKAKGPLASQISGLYLPYKVLPGVDEVVGVGIVGYSKETPFLSNPGTNLVFEDEITAL 200

QPEVDKLLKTLNVNKIIALGHSGFETDKLIAQKVRGVDVVVGGHSNTFLYTGNNPSKEVPAGKYPFIVTSDDRKVPVQYAYAFGKYLGYLKIEFDERGNV
 210     220     230     240     250     260     270     280     290     300
Rh_CD73L_Prot QPEVDKLLKTLNVNKIIALGHSGFETDKLIAQKVRGVDVVVGGHSNTFLYTGNNPSKEVPAGKYPFIVTSDDRKVPVQYAYAFGKYLGYLKIEFDERGNV 300
Cyno-CD73_Pro QPEVDKLLKTLNVNKIIALGHSGFETDKLIAQKVRGVDVVVGGHSNTFLYTGNNPSKEVPAGKYPFIVTSDDRKVPVQYAYAFGKYLGYLKIEFDERGNV 300
Hu_CD73-L_Pro QPEVDKLLKTLNVNKIIALGHSGFEMDKLIAQKVRGVDVVVGGHSNTFLYTGNNPSKEVPAGKYPFIVTSDDRKVPVQYAYAFGKYLGYLKIEFDERGNV 300

ISSHGNPILLNSSIPEDPSIKADINKWRIKLDNYSTQELGKTIYVLDGSSQSCRFRECNMGNLICDAMINNNLRHADEMFWNHVSMCILNGGGIRSPIDE
 310     320     330     340     350     360     370     380     390     400
Rh_CD73L_Prot ISSHGNPILLNSSIPEDPSIKADINKWRIKLDNYSTQELGKTIYVLDGSSQSCRFRECNMGNLICDAMINNNLRHADEMFWNHVSMCILNGGGIRSPIDE 400
Cyno-CD73_Pro ISSHGNPILLNSSIPEDPSIKADINKWRIKLDNYSTQELGKTIYVLDGSSQSCRFRECNMGNLICDAMINNNLRHADEMFWNHVSMCILNGGGIRSPIDE 400
Hu_CD73-L_Pro ISSHGNPILLNSSIPEDPSIKADINKWRIKLDNYSTQELGKTIYVLDGSSQSCRFRECNMGNLICDAMINNNLRHADETFWNHVSMLCINNGGGIRSPIDE 400

RNNGITWENLAAVLPFGGTFDLVQLKGSTLKKAFEHSVHRYGQSTGEFLQVGGIHVVYDLSRKPGRVVKLDVLCCKRVPVSYDPLKMDIYKVIPLPNF
 410     420     430     440     450     460     470     480     490     500
Rh_CD73L_Prot RNNGITWENLAAVLPFGGTFDLVQLKGSTLKKAFEHSVHRYGQSTGEFLQVGGIHVVYDLSRKPGRVVKLDVLCCKRVPVSYDPLKMDIYKVIPLPNF 500
Cyno-CD73_Pro RNNGITWENLAAVLPFGGTFDLVQLKGSTLKKAFEHSVHRYGQSTGEFLQVGGIHVVYDLSRKPGRVVKLDVLCCKRVPVSYDPLKMDIYKVIPLPNF 500
Hu_CD73-L_Pro RNNGITWENLAAVLPFGGTFDLVQLKGSTLKKAFEHSVHRYGQSTGEFLQVGGIHVVYDLSRKPGRVVKLDVLCCKRVPVSYDPLKMDIYKVIPLPNF 500

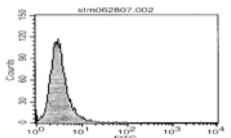
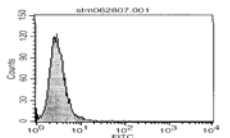
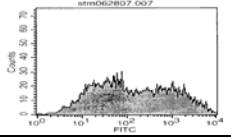
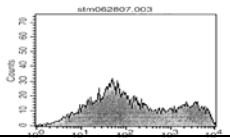
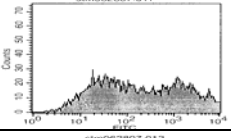
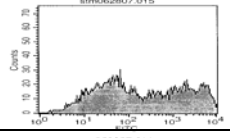
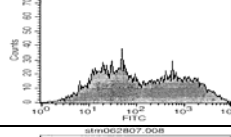
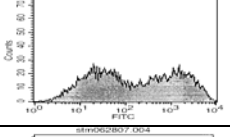
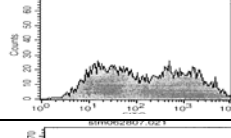
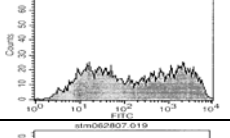
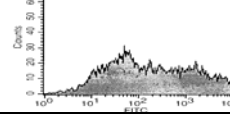
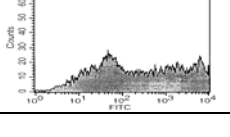
LANGGDFQMIKDELLRHDSGDQDINVVSTYISKMKVIYPAVEGRIFSTGSHCHGFSFLIFLSFCVAVIFVLYQ
 510     520     530     540     550     560     570
Rh_CD73L_Prot LANGGDFQMIKDELLRHDSGDQDINVVSTYISKMKVIYPAVEGRIFSTGSHCHGFSFLIFLSFCVAVIFVLYQ 574
Cyno-CD73_Pro LANGGDFQMIKDELLRHDSGDQDINVVSTYISKMKVIYPAVEGRIFSTGSHCHGFSFLIFLSFCVAVIFVLYQ 574
Hu_CD73-L_Pro LANGGDFQMIKDELLRHDSGDQDINVVSTYISKMKVIYPAVEGRIFSTGSHCHGFSFLIFLSLWAVIFVLYQ 574

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Part II. Anti-CD73 antibody reactivity with rhesus CD73

Method: 293 T cells were transfected with human or rhesus CD73 cDNA and cell surface expression was detected using anti-human CD73 antibodies. All antibodies were clustered as anti-human CD73 in the 4th and 5th Leucocyte Typing workshops.

Antibodies tested on transfected 293T cells:

Clone	Leucocyte Typing Workshop Designation	Commercial Source	Human CD73	Rhesus CD73
Neg control	n/a	n/a		
AD2	Leucocyte Typing IV B43	BD Bioscience		
7G2	Leucocyte Typing IV B124	Invitrogen		
1E9	Leucocyte Typing IV B121	Santa Cruz Biotechnology		
5N4	Leucocyte Typing V CD73.6	n/a		
5N2	Leucocyte Typing V CD73.5	n/a		

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