

# SEQUENCE LISTING

<110> Centro de Investigación y de Estudios Avanzados del  
Instituto Politécnico Nacional

<120> Genetic system for the production of leupeptin and its use for  
heterologous protein production

<130> Cinvestav31

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<170> PatentIn version 3.5

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Ala Cys Ala Gln Gly Leu Val Asp Arg His Glu Ala Phe Thr Leu Arg
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Thr Arg Phe Val Leu Arg Asp Gly Glu Leu Trp Gln His Ile Asp Thr
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Glu Ala Ala Leu Val Trp Ser Ser Ala Val Ala His Gly Ala Gly Asp
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Val Glu Glu Trp Met Ala Ala Glu His His Arg Pro Phe Asp Leu Gly
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Ser Gly Pro Leu Phe Arg Ala Ala Phe Thr Leu Leu His Thr Pro Glu
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Gly Ala Val Leu Ala Leu Ala Met His His Ile Val Ala Asp Gly Trp
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Ser Val Gly Ile Leu Val Arg Glu Leu Leu Thr Ala Tyr Ala His Tyr
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Gly Asp Gly Pro Asp Gly Gly Pro Asp Gly Thr Asp Arg Arg Arg Pro
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Ala Gly Phe Val Phe Thr Ala Pro Glu Ala Pro Glu Tyr Gln Tyr Ala
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Asp Phe Ser Asp Trp Gln Glu Glu Trp Leu Arg Gly Pro Ala Ala Gln

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Arg Ala Leu Pro Asp Gly	Asp Leu Glu Tyr Leu Gly	Arg Ile Asp His		
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Gln Val Lys Leu Arg Gly	Phe Arg Ile Glu Leu Gly	Glu Val Glu Ala		
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Ala Leu Ala Ala His Pro	Leu Val Asp Ala Ala Val	Ala Leu Val Ala		
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Phe Thr Pro Asp Pro Ala	Gly Asn Asp Val Leu Val	Gly Tyr Leu Ala		
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Val Thr Gly Ala Glu Asp	Gly Ala Gly Pro Thr Val	Glu Glu Met Arg		
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Glu His Leu Ala Arg Arg	Leu Pro Gly Tyr Met Ile	Pro Gly Ala Phe		
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Val Thr Leu Pro Ala Phe	Pro Leu Thr Ala Asn Gly	Phe Thr Lys Thr		
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Leu Ala Arg Pro Ala Ala	Thr Gly Gly Pro Ala	Phe Thr Arg Met		
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Ser Trp Tyr Leu Ala Pro	Leu Pro Gly Glu Leu	His Asp Arg Leu		
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Glu Thr Asp Gly Asp Lys	Val Val Gly Val Phe	Leu Asn Thr Leu		
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Val Thr Gly Glu Leu Phe	Leu Glu Gly Val Gly Phe	Thr Leu Ala		
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Asp Gly Tyr His Gly Arg	Pro Glu Leu Thr Ala Glu	Arg Phe Ile		
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Glu His Thr Gly Pro Asp	Gly Thr Leu Arg Arg Leu	Tyr Arg Thr		
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Gly Asp Leu Ala Arg His	Arg Pro Asp Gly Ala Leu	Glu Tyr Ala		
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Arg Glu Arg Leu Pro Glu	Tyr Met Val Pro Ser Ala	Trp Val Cys		
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Ala Leu Pro Asp Pro Asp	Pro Ala Asp Leu Arg Gly	Gly Val Glu		



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 <221> MISC\_FEATURE  
 <223> Protein coded by the LeupB gene from the clone 9\_18N  
  
 <220>  
 <221> MISC\_FEATURE  
 <223> Argininosuccinate lyase (EC 4.3.2.1)  
  
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 Arg Gly Gly Pro Ala Glu Leu Leu His Asp Glu Val Leu Ala Pro Gln  
 20                      25                      30  
 Phe Gly Phe Glu Ser Arg His Leu Leu Arg His Tyr Phe Thr Val Ala  
 35                      40                      45  
 Ile Glu Lys Thr Leu Ala Ala Glu Tyr Val Arg Met Asp Leu Ile Thr  
 50                      55                      60  
 Glu Glu Glu Ala His Arg Ile Ala Ala Leu Leu Asp Ala Val Gly Pro  
 65                      70                      75                      80  
 Asp Thr Leu Ser Ala Gln Pro Gly Ala Asn Met Ser Asp Ile Ala Phe  
 85                      90                      95  
 Ala Leu Glu Arg His Val Glu Ala Phe Thr Gly Leu Pro Ala Pro Val  
 100                      105                      110  
 Thr His Trp His Ala Asp Arg Ser Arg Asn Asp Leu Gln Ala Cys Ala  
 115                      120                      125  
 Gln Val Met Tyr Gly Arg Asp Gln Leu Ala Arg Phe Ala Ser Ala Leu  
 130                      135                      140  
 Leu Glu Leu Gly Ser Ile Val Val Glu Leu Ala Gln Gln Thr Cys Asp  
 145                      150                      155                      160  
 Leu Pro Met Pro Phe Thr Gly Tyr Thr His Phe Gln Ala Ala Gln Ile

				165						170						175			
Ile	Thr	Pro	Gly	Phe	His	Leu	Ala	Ala	Leu	Ser	Glu	His	Leu	Leu	His				
			180					185					190						
Thr	His	Ala	Arg	Leu	Leu	Asn	Ala	Tyr	Asp	Gly	Ile	Asp	Ala	Cys	Pro				
		195					200					205							
Leu	Gly	Ala	Gly	Ala	Met	Ala	Gly	Gln	Glu	Leu	Pro	Trp	Asp	Arg	Asp				
	210					215					220								
Phe	Thr	Arg	Met	Ala	Arg	Leu	Leu	Gly	Phe	Ser	Arg	Pro	Gln	Pro	His				
225					230					235					240				
Ala	Leu	Thr	Ala	Val	Ala	Ser	Arg	Arg	Trp	Ser	Ala	Glu	Leu	Thr	Ala				
				245					250					255					
Glu	Leu	Ser	Leu	Leu	Gly	Thr	Ala	Leu	Ser	Arg	Phe	Thr	Thr	Asp	Leu				
			260					265					270						
Leu	Thr	Trp	Gly	Gly	Ser	Glu	Tyr	Gly	Phe	Ile	Glu	Phe	Thr	Leu	Pro				
		275					280					285							
Asp	Glu	Leu	Ser	Gly	Ile	Ser	Ser	Ala	Met	Pro	Gln	Lys	Lys	Asn	Tyr				
	290					295					300								
Pro	Val	Leu	Glu	Arg	Ile	Arg	Gly	Arg	Thr	Ala	His	Leu	Thr	Ala	Phe				
305					310					315					320				
His	Phe	Asp	Val	Leu	Leu	Gly	Gln	Arg	Asn	Thr	Pro	Phe	Cys	Asn	Leu				
				325					330					335					
Val	Glu	Val	Ser	Lys	Glu	Ala	Gly	Phe	Thr	Thr	His	Leu	Leu	Asn	Ala				
			340					345					350						
Phe	Asp	Ser	Ala	His	Gly	Thr	Val	Arg	Leu	Leu	Thr	Glu	Val	Leu	Arg				
		355					360					365							
Arg	Leu	Thr	Phe	Arg	Ala	Asp	Arg	Met	Arg	Gln	Val	Cys	Glu	Arg	Glu				
	370					375					380								
Phe	Leu	Gly	Gly	Phe	Ser	Leu	Ala	Asn	Ala	Leu	Cys	Leu	Thr	Glu	Gly				
385					390					395					400				
Val	Pro	Trp	Arg	Phe	Thr	Thr	Ala	Gln	Val	Val	Ala	Gly	Lys	Tyr	Val				
				405					410					415					
Val	Leu	Ala	Ala	Thr	Ala	Gly	Ala	Ala	Pro	Ala	Pro	Gly	Glu	Pro	Ala				
			420					425					430						
Leu	Leu	Ala	Glu	Ala	Ala	Ala	Gly	His	Gly	Ile	Thr	Leu	Ala	Asp	Pro				
		435					440					445							
Ala	Arg	Leu	Leu	Asp	Glu	Ala	Phe	Asp	Val	Asp	Arg	Gly	Leu	Glu	Arg				
	450					455					460								
Phe	Thr	Met	Val	Ser	Ala	Gly	Ser	Ala	Arg	Pro	Asp	Ala	Val	Arg	Ala				
465					470					475					480				
Val	Leu	Arg	Asp	Gln	Gln	Glu	Thr	Tyr	Glu	Gln	Leu	Arg	Ala	Asp	Trp				
				485					490					495					
Gln	Ala	Arg	Ala	Asp	Ala	Leu	Arg	Ala	Gly	Ala	Glu	Glu	Gly	Glu	Arg				

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Ala Leu Tyr Gly Thr Pro Ala Asp Lys Thr Gln Glu Phe Thr Glu Asp		
515	520	525
Gly Asp Gly Thr Arg Thr Arg Val Gln His Ala His		
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<210> 12  
 <211> 336  
 <212> PRT  
 <213> Streptomyces roseous  
  
 <220>  
 <221> MISC\_FEATURE  
 <223> Protein coded by the LeupC gene from the clone 9\_18N  
  
 <220>  
 <221> MISC\_FEATURE  
 <223> Threonine kinase  
  
 <400> 12

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Thr Thr Ala Val Pro Val Gly Pro Gly Ser Gly Ser Ala Phe Gly Thr
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Phe Gly Glu Leu Leu Gln Gly Ala Leu Pro His Pro Phe Thr Gln Gly
35 40 45
Asp Phe Leu Val Thr Phe Pro Leu Ala Arg Trp Ala Thr Ala Val Phe
50 55 60
His Pro Glu Pro Gly Arg Arg Glu Val Arg Val Arg Pro Ala His Lys
65 70 75 80
Ala Lys Ser Arg Arg Val Ala Glu Ala Val Leu Ala Ala Leu Gly Thr
85 90 95
Ala Gly Gly Gly Leu Leu Glu Val Phe Thr Ala Gly Asp Leu Pro Glu
100 105 110
Gly Lys Gly Leu Ala Ser Ser Ser Ala Asp Leu Val Ala Thr Val Arg
115 120 125
Ala Val Gly Ala Ala His Gly Arg Ala Phe Thr Pro Asp Glu Thr Glu
130 135 140
Gly Phe Leu Arg Gly Ile Glu Pro Ala Asp Gly Val Met Tyr Asp Glu
145 150 155 160
Ile Val Ala Phe Phe Thr His His Arg Glu Val Arg Leu Gly Arg Arg
165 170 175
Leu Gly Val Leu Pro Pro Leu Thr Val Val Ala His Asp Glu Gly Gly
180 185 190
Gln Val Asp Thr Val Ala His Asn Arg Gly Ala Arg Ala Ile Asp Ala
195 200 205
Ala Asp Arg Glu Glu Tyr Ala Arg Leu Leu Asp Arg Leu Thr Asp Ala

210		215		220
Phe Thr Val Ala Arg Gly Asp Leu Ala Glu Val Gly Ala Val Ala Thr				
225		230		235
Arg Ser Ala Glu Met Asn Ala Arg Arg Arg Arg Ala Gly Phe Ala				
	245		250	255
Glu Leu His Ala Leu Cys Arg Glu Val Asp Gly Leu Gly Leu Val Leu				
	260		265	270
Ala His Ser Gly Thr Met Leu Gly Val Leu Leu Glu Phe Thr Ala Asn				
	275		280	285
Asp Pro Ala Leu Ala Gly Lys Thr Glu His Ile Arg Ala Gly Cys Ala				
	290		295	300
Ala Leu Gly Gly Glu Val Ser Val His Arg Ser Leu Gly Ala Asp Asp				
305		310		315
Ser Trp Ser Pro Gln Ala Pro Pro Ala His Pro Thr Glu Leu Glu Ile				
	325		330	335

<210> 13  
 <211> 781  
 <212> PRT  
 <213> Streptomyces roseous

<220>  
 <221> MISC\_FEATURE  
 <223> Protein coded by the LeupD gene from the clone 9\_18N

<220>  
 <221> MISC\_FEATURE  
 <223> Cysteine synthase (EC 2.5.1.47)

<400> 13

Met Leu Phe Asp Thr Val Thr Asp Ala Ile Gly Gly Thr Pro Leu Val				
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Arg Leu Arg Leu Gly Glu Ala Arg Gly Val Glu Val Tyr Ala Lys Leu				
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Glu Leu Gln Asn Leu Phe Ala Met Lys Asp Arg Val Phe Thr Ala Arg				
	35		40	45
Asn Ile Leu Leu Glu Ala Arg Arg Leu Gly Thr Leu Lys Pro Gly Asp				
	50		55	60
Pro Val Ile Glu Ser Ser Ser Gly Thr Met Ala Leu Gly Val Ala Leu				
65		70		75
Val Gly Arg Ser Leu Gly His Glu Val His Ile Val Thr Asp Pro Arg				
	85		90	95
Ile Asp Pro Val Thr Leu Ala Lys Phe Thr Leu Arg Ala Leu Gly Cys				
	100		105	110
Arg Val His Ile Val Glu Ala Met Thr Ser His Gly Trp Gln Ser Ala				
	115		120	125
Arg Leu Glu Arg Leu Ala Glu Leu Leu Asp Glu Leu Pro Gly Ala Phe				

130						135						140				
Trp	Pro	Gln	Gln	Tyr	Thr	Asn	Pro	Asp	Asn	Pro	Gly	Ala	Tyr	Arg	Pro	
145					150					155					160	
Leu	Ala	Gly	Glu	Phe	Thr	Leu	Leu	Ala	Asp	Leu	Gly	Arg	Phe	Asp	Thr	
				165					170					175		
Leu	Val	Gly	Ala	Val	Gly	Ser	Gly	Gly	Ser	Leu	Cys	Gly	Thr	Ala	Arg	
			180					185					190			
Ala	Leu	Arg	Glu	Arg	Leu	Pro	Ala	Leu	His	Val	Val	Gly	Val	Asp	Cys	
		195					200					205				
Val	Gly	Ser	Ala	Leu	Phe	Gly	Gln	Pro	Asp	Val	Pro	Gln	Arg	Leu	Gln	
	210					215					220					
Phe	Thr	Ser	Gly	Leu	Gly	Asn	Ser	Leu	Leu	Pro	Lys	Asn	Leu	Asp	Arg	
225					230					235					240	
Thr	Leu	Val	Asp	Glu	Val	His	Trp	Leu	Asn	Asp	His	Glu	Ala	Phe	Ala	
				245					250					255		
Ala	Thr	Trp	Asp	Leu	Ala	Arg	Glu	Gln	Gln	Ile	Phe	Gly	Gly	Asn	Thr	
			260					265					270			
Ser	Gly	Ser	Val	Tyr	Arg	Val	Leu	Thr	Gly	Leu	Ala	Phe	Thr	Asp	Gly	
		275					280					285				
Ala	Glu	Pro	Gly	Thr	Arg	Ile	Val	Gly	Ile	Leu	Pro	Asp	Arg	Gly	Asp	
	290					295					300					
Arg	Tyr	Ala	Asp	Thr	Val	Tyr	Asn	Asp	Glu	His	Trp	Asp	Ala	Asn	Arg	
305					310					315					320	
Leu	Arg	Glu	Val	Pro	Thr	Ala	Thr	Ala	Pro	Ala	Ala	Leu	Gly	Pro	Asp	
				325					330					335		
Gly	Thr	Ala	Arg	Thr	Trp	Ser	Thr	Phe	Thr	Val	Ala	Tyr	Ser	Pro	Pro	
		340						345					350			
Ala	Gly	Ile	Gly	Arg	His	Leu	Leu	Phe	Val	Glu	Ser	Asn	Thr	Thr	Gly	
		355					360					365				
Thr	Gly	Met	Leu	Ala	Leu	Ala	Arg	Ala	Arg	Glu	Leu	Gly	Thr	Val	Pro	
	370					375					380					
Val	Leu	Leu	Thr	Gly	Asp	Pro	Asp	Arg	Tyr	Arg	Gly	Leu	Ala	Glu	Ser	
385					390					395					400	
Gly	Ala	Glu	Val	Phe	Thr	Val	Arg	Cys	Asp	Thr	Asn	Ser	Asp	Ala	Ala	
				405					410					415		
Leu	Arg	Ala	Ala	Val	Gln	Glu	Arg	Phe	Arg	Arg	Glu	Glu	Ile	Ala	Gly	
				420				425					430			
Val	Thr	Thr	Thr	Ser	Asp	Phe	Tyr	Val	Pro	Ala	Ala	Ala	Arg	Ile	Ala	
		435					440					445				
Gln	Trp	Leu	Gly	Leu	Pro	Gly	Asn	Ala	Pro	Glu	Ala	Val	Ala	Val	Cys	
	450					455					460					
Phe	Thr	Arg	Asp	Lys	Ser	Ala	Leu	Arg	Glu	Arg	Leu	Arg	Ala	Ala	Gly	

465		470		475		480
Val Arg Gln Pro Arg Tyr Ser Leu Val Arg Glu Pro Ala Gly Ala Ala	485		490		495	
Ala Ala Val Ala Arg Thr Gly Leu Pro Cys Val Val Lys Pro Ala Asp	500		505		510	
Asp Ser Gly Ser Thr Asn Val Leu Leu Cys Ala Asp Phe Thr Glu Ala	515		520		525	
Glu Ala Arg Ala Gln Ile Gln Arg Ile Leu Ala Ile Asp Thr Asn Val	530		535		540	
Arg Gly Met Pro Thr Ala Arg Thr Val Leu Val Glu Glu Tyr Leu Asp	545		550		555	560
Gly Pro Glu Tyr Ser Val Glu Met Phe Gly Leu Asp Gly Gln Ala Val	565		570		575	
Cys Val Gly Ile Thr Ala Lys Ser Phe Thr Val Thr Ala Gly Pro His	580		585		590	
Phe Val Glu His Arg His Leu Phe Pro Ala Pro Leu Pro Ala Ala Thr	595		600		605	
Ala Gln Leu Ile Thr Asp Thr Val Thr Ala Ala Leu Asp Ala Ala Gly	610		615		620	
Ile Arg Leu Gly Ala Thr His Thr Glu Val Lys Leu Thr Ala Asp Gly	625		630		635	640
Pro Ala Leu Val Phe Thr Glu Ile Asn Pro Arg Pro Ala Gly Gly Met	645		650		655	
Ile Pro Glu Leu Ile Arg Leu Ala Thr Gly Val Asp Leu Leu Asp Ala	660		665		670	
Gln Leu Arg Ala Ala Leu Gly Leu Pro Pro His Leu Lys Ala Glu Glu	675		680		685	
Ala Gly His Ala Gly Ile Gln Phe Leu Leu Ala Asp Thr Asp Gly Thr	690		695		700	
Phe Thr Leu Thr Ala Val His Gly Ala Glu Ala Ala Ala Val Val Glu	705		710		715	720
Gly Val Glu Ser Val Leu Val Thr Ala Ala Pro Gly Thr Pro Val Arg	725		730		735	
Arg Pro Arg Ser Ala Ser Asp Arg Leu Gly His Val Ile Ala Arg His	740		745		750	
Ala Glu Pro Asp Gly Val His Thr Ala Leu Asp Ala Phe Thr Ala Arg	755		760		765	
Ala Leu Val His Leu Asp Ile Gln Ala Ala Pro Arg Ser	770		775		780	