Legends of the supplementary figures

Figure S1. The mRNA secondary structure near the start codon AUG predicted using Centroidfold algorithm. The mRNA secondary structures of the expressed genes are presented on the left and those of the unexpressed genes on the right. Green arrows indicate the first position of the start codon AUG (nt 44-46); black arrows indicate the first position of the SD sequence (nt 29-35). The overall free energy of each secondary structure (kcal/mol) is shown in between brackets.

Figure S2. Screen-shots of the Rchange output files of p146, NcoI-E2s, HBsAg-1 and HBsAg-2. The output data are presented as follows: Sequence positions of mutations (mut_pos1, mut_pos2) are in 0-based indexing convention. "mutation_data" is semicolon (';') delimited entries representing different mutations to the original base(s). For each mutation, there are 4 fields delimited by comma ',' representing mutated base(s), entropy change, internal energy change, Helmholtz free energy change.

The data of the mutated position are highlighted in yellow. For, each position, the highest free energy increase induced by a single mutation is highlighted in green while the energy increases induced by the other possible substitutions are highlighted in red.

Figure S3. The predicted mRNA secondary structures near the start codons of the mutated p146, NcoI-E2s, HBsAg-1 and HBsAg-2 genes shows that the start codon AUG (red arrows) and the SD sequence (black arrows) are exposed from the stable stem-loop structural context.

Table S1. Amino acid composition of HEV and HBV target proteins

AA	HEV target proteins		HBV target proteins	
	Avg.	S.D.	Avg.	S.D.
Phe	5.00	1.41	13.67	3.51
Leu	15.88	2.75	29.33	6.03
Ile	6.00	1.41	11.00	3.00
Val	15.38	1.92	9.33	0.58
Ser	18.88	2.42	20.33	5.51
Pro	11.00	2.62	20.00	4.58
Thr	20.25	1.98	15.33	4.62
Ala	16.13	3.04	5.33	1.15
Tyr	10.38	1.30	4.00	1.00
His	3.50	2.33	0.67	0.58
Gln	7.63	2.56	5.67	2.31
Asn	7.50	0.53	3.67	1.53
Lys	4.88	1.36	2.00	0.00
Asp	10.13	4.19	2.67	0.58
Glu	5.25	2.43	1.67	0.58
Cys	3.00	1.41	12.33	2.08
Arg	8.25	2.43	4.67	2.31
Gly	12.50	2.83	11.67	4.04
Met	2.75	1.39	5.67	0.58
Trp	3.00	0.00	11.67	1.53

HEV target proteins: p166, p179, p216, p222, p231, p146 and E2s
HBV target proteins: HBsAg-1, HBsAg-2 and HBsAg-3
AA: amino acids
Avg.: average; S.D.: standard deviation

Table S2. Relative synonymous codon usage (RSCU) in HEV and HBV target genes

AA[a]	CODONS	HEV genes Avg.[b]	S.D.[c]	HBV genes Avg.	S.D.	AA	CODONS	HEV genes Avg.	S.D.	HBV genes Avg.	S.D.
Phe	TTT	**80.0**	5.5	39.0	3.0	Ala	GCT	**55.5**	4.3	**38.8**	9.8
	TTC	20.0	5.5	**61.0**	3.0		GCC	25.3	2.0	19.5	4.8
Leu	TTA	0.0	0.0	10.3	3.3		GCA	16.8	2.8	30.5	4.8
	TTG	15.8	4.7	17.0	0.3		GCG	2.5	2.8	11.3	9.8
	CTT	**42.5**	2.2	16.5	4.5	Tyr	TAT	**64.5**	2.5	44.0	21.0
	CTC	14.8	1.3	15.3	5.8		TAC	35.5	2.5	**56.0**	21.0
	CTA	6.0	0.8	**22.7**	0.8	His	CAT	**98.0**	5.0	0.0	0.0
	CTG	21.0	4.0	18.2	1.7		CAC	2.0	5.0	**66.5**	57.5
Ile	ATT	37.3	4.3	28.0	9.7	Gln	CAA	16.5	9.0	**51.0**	13.5
	ATC	**61.0**	4.7	**42.0**	4.0		CAG	**84.0**	9.0	49.0	13.5
	ATA	2.0	4.3	30.0	5.7	Asn	AAT	**87.0**	1.0	45.0	39.5
Val	GTT	**48.3**	3.5	**43.0**	2.5		AAC	13.0	1.0	**55.0**	39.5
	GTC	25.8	3.0	21.5	1.3	Lys	AAA	30.0	10.0	**50.0**	0.0
	GTA	6.3	0.5	17.8	6.0		AAG	**70.0**	10.0	**50.0**	0.0
	GTG	20.0	4.3	17.8	6.0	Asp	GAT	**70.0**	8.0	0.0	0.0
Ser	TCT	**61.0**	7.8	16.2	1.5		GAC	30.0	8.0	**100.0**	0.0
	TCC	17.8	2.2	21.3	0.5	Glu	GAA	0.0	0.0	0.0	0.0
	TCA	6.8	2.7	**26.5**	1.8		GAG	**100.0**	0.0	**100.0**	0.0
	TCG	5.2	0.5	10.8	3.2	Cys	TGT	42.5	10.0	**58.5**	7.5
	AGT	5.2	0.5	22.2	5.7		TGC	**57.5**	10.0	41.5	7.5
	AGC	4.2	10.2	2.8	2.5	Arg	CGT	38.3	7.2	**38.8**	9.7
Pro	CCT	**67.3**	2.0	28.3	1.3		CGC	**43.8**	6.2	11.2	9.7
	CCC	13.3	6.5	30.3	2.5		CGA	3.0	4.8	0.0	0.0
	CCA	18.0	3.0	**41.5**	1.5		CGG	14.7	3.0	11.2	9.7
	CCG	1.8	4.5	0.0	0.0		AGA	0.0	0.0	**38.8**	9.7
Thr	ACT	**80.3**	2.3	24.8	4.5		AGG	0.0	0.0	0.0	0.0
	ACC	10.3	2.8	**28.5**	1.3	Gly	GGT	30.0	1.8	9.5	4.0
	ACA	4.8	0.5	**28.8**	7.8		GGC	**41.3**	1.3	9.5	8.3
	ACG	4.8	0.5	17.8	2.0		GGA	2.0	4.8	**52.5**	4.3
							GGG	27.0	4.8	28.5	0.0

The values are expressed as percentage

The preferentially used codons for each amino acid are indicated in bold.

[a] amino acids; [b] Average; [c] standard deviation.

Table S3.

Genes	Sequences of the -43/ATG/+43 mRNA segments
NcoI-E2s	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>UCCCCUGCUCCUUCUCGCCCUUUUUCU GUGCUUCGUGCUAAUG
Mutated NcoI-E2s	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>UCCCCUG==AU==G==A==UUCUCGCCCUUUUUCU GUGCUUCGUGCUAAUG
p146	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>GCUCCUUCUCGCCCUUUUUCUGUGCUU CGUGCUAAUGAUGUGC
Mutated p146	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>G==A==UG==A==UUCUCGCCCUUUUUCUGUGCU UCGUGCUAAUGAUGUGC
HBsAg-1	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>GAGAGCACAACAUCAGGAUUCCUAGG ACCCCUGCUCGUGUUAC
Mutated HBsAg-1	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>GAGAGCACAACAUCAG==AT==UUCCUAGGA CCCCUGCUCGUGUUAC
HBsAg-2	CCUCUAGAAAUAAUUUUGUUUAACUUUA==AGAAGGA==G AUAUACC<mark>AUG</mark>GCUCUGCUGCUAUGCCUCAUCUUCUUG UUGGUUCUUCUGGACU
Mutated HBsAg-2	CCTCTAGAAATAATTTTGTTTAACTTTA==AGAAGGA==GAT ATACC<mark>ATG</mark>GCTCTGCT==T==CT==G==TGCCTCATCTTCTTGTTGG TTCTTCTGGACT

The SD sequences are highlighted in green

The start codons are highlighted in yellow

The substitutions in the mutated genes are highlighted in cyan