

# CTF4, a chicken transcription factor of the helix-loop-helix class A family

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The cDNA for a new member of the family of basic Helix-Loop-Helix (bHLH) proteins was isolated from a chick embryo cDNA expression library using the strategy of Singh *et al.* (1). Screening was performed with oligomers of the chick muscle acetylcholine receptor  $\alpha$ - and  $\delta$ -subunit enhancers (2, 3). Binding specificity for the E-box motif was established by gel shift competition with wildtype and mutant  $\alpha$ -subunit enhancers.

We have sequenced 2061 bp of the clone which has the 1569-bp (523 amino acids) open reading frame shown in Figure 1. Protein alignment analysis reveals that a 286-residue HLH-containing fragment is highly similar to a segment in several regulatory proteins: HTF4 (82%), ITF2 (63%), SEF21B (63%) and E12-related proteins (47%). We conclude that the gene is the chicken homolog of the human class A bHLH protein HTF4 recently described by Zhang *et al.* (4) and propose the designation CTF4.

Transfection of this cDNA into NIH3T3 cells indicates that the factor which it encodes activates transcription of a cotransfected reporter gene containing an oligomerized E-box motif. Effects of cotransfection of this cDNA and myogenic factors are under investigation.

## REFERENCES

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C	MSSFHRGSTS	SSPYVAASHT	FPVNGSENIL	GNRNGAGGS	QTGDALGKAL	50
C	ASIYSPDHTS	<b>SSFFSNPSTP</b>	<b>VGSPSPLTGA</b>	<b>SQWSRSGGQA</b>	<b>PSSPMYENSL</b>	100
H		** *****	*****	** * ****	*****	
		<b>TS</b>	<b>SSFFSNPSTP</b>	<b>VGSPSPLTGT</b>	<b>SQWPRPGGQA</b>	<b>PSSPSYENSL</b>
C	<b>HSLKNRVEEQ</b>	<b>LHEHLQDAMS</b>	<b>FLKDVCESR</b>	<b>MEDRLDRLDD</b>	<b>AIHVLRNHAV</b>	150
H	<b>HSL</b>	-----	-----	<b>QSR</b>	<b>MEDRLDRLDD</b>	<b>AIHVLRNHAV</b>
C	<b>GPSTSLSGGH</b>	<b>GDIHSLGGS</b>	<b>HMGPIGSLNS</b>	<b>SYGASSLVAA</b>	<b>NRQASIVMAG</b>	200
H	<b>GPSTSLPAGH</b>	<b>SDIHSLLGGS</b>	<b>HMAPIGSLNS</b>	<b>NYGGSBLVAS</b>	<b>SRSASMVGT-</b>	
C	<b>SSGGKRQLNS</b>	<b>NEAVLPSTVS</b>	<b>AQTTELNHKT</b>	<b>QESYRALSOG</b>	<b>LQSQSVAIGP</b>	250
H	<b>HREDSVSLNG</b>	<b>NESVLSSTVT</b>	<b>TSSTDLNHKT</b>	<b>QENYR---</b>	<b>GG LQSQSGTVVT</b>	
C	<b>TEIKSEHKKE</b>	<b>DENIHEPPSS</b>	<b>DDMKSDEESS</b>	<b>QKDIKVSRRG</b>	<b>RTSSTNEDED</b>	300
H	<b>TEIKTENKEK</b>	<b>DENIHEPPSS</b>	<b>DDMKSDEESS</b>	<b>QKDIKVSRRG</b>	<b>RTSSTNEDED</b>	
C	<b>LNPEQKIERE</b>	<b>KERRMANNAR</b>	<b>ERLRYRDINE</b>	<b>AFKELGRMCO</b>	<b>LHLKSEKPT</b>	350
H	<b>LNPEQKIERE</b>	<b>KERRMANNAR</b>	<b>ERLRYRDINE</b>	<b>AFKELGRMCO</b>	<b>LHLKSEKPT</b>	
C	<b>KLILILHOAVA</b>	<b>VILSLEQOVR</b>	<b>ERNLNPKAAC</b>	<b>LKRREEKYS</b>	<b>AVVRRQHHTQ</b>	400
H	<b>KLILILHOAVA</b>	<b>VILSLEQOVR</b>	<b>ERNLNPKAAC</b>	<b>LKRREEKYS</b>	<b>AVSASPPTTL</b>	
C	<b>EAT-LALSET</b>	<b>TNFMVICEHQ</b>	<b>PELLVGYTEG</b>	<b>DLSSQHRQL</b>	<b>TLYEDTNLTG</b>	449
H	<b>PGTHPGLSET</b>	<b>TNFMGHM</b>				
C	<b>ENTCKQETQM</b>	<b>QSYDQSYWST</b>	<b>PASGLKIEDL</b>	<b>QIEFQPTKYS</b>	<b>EHILLQAVC</b>	499
C	<b>RFCTIRDCCD</b>	<b>LSTHCGSLV</b>	<b>PKNL</b>			523

Figure 1. Amino acid sequence corresponding to the cDNA that encodes CTF4. The chicken gene sequence (C) is aligned with the recently published human gene HTF4 (H). Residues common to both proteins are bolded and highlighted with asterisks; the bHLH region and the adjacent loop 2 and helix 3 characteristic of class A family proteins are underlined. Numbers refer to position in the chicken sequence.

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