

cDNA and deduced amino acid sequence of a novel cytochrome P-450 from female rat liver mRNA with high homology to P-450 IIC family

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Liver mRNA from female adult rats which had been maintained on a cholestyramine diet (4% w/w) was used to construct a cDNA-library in λgt11. The library was screened with a 1233 bp BamHI/XbaI fragment of a cDNA-clone for the male-specific P-450 IIC11 [P-450 M-1, P-450 16α] (1) and many clones hybridised with the probe. Dot blot analysis with specific probes for other members of the P-450 IIC family allowed identification of most of the positive clones as either P-450 PB1, P-450 f, or P-450 15β. All these species are considered members of the P-450 IIC family. In addition a clone for P-450 IIE1 (P-450 j) was also identified. One clone that could not be identified was isolated, sequenced and designated P-450 cl17. The deduced amino acid sequence has the highest homology with members of the rat P-450 IIC family P-450 M-1 (59%), P-450 PB1 (58%), P-450 f (56%), P-450 g and P-450 15β (53%) and therefore we suggest that this novel species should be classified as a new member of this family. A putative heme-binding region can be identified in this sequence (see figure). In mRNA samples from both male and female adult rat liver, P-450 cl17 hybridised to a species of apparently 2000 bp and this is consistent with the size of a mRNA for a P-450 species (results not shown).

REFERENCES

1. Yoshioka,H. *et al.* (1987) *J. Biol. Chem.* **262**, 1706–1711.
2. Gonzalez,F.J. (1989) *Pharmacol. Rev.* **40**, 243–288.

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GAGGGCTAAAATGGAGCTGCTGGGATTCACTACGCTTGCTCTTGTGGTTTCGGTGACTTGCCTCAGCCTCCTTTCAGTGTGGACAAAACTGCGCACTAGGGGGAGGCTGCCCCCG  115
          MetGluLeuLeuGlyPheThrThrLeuAlaLeuValValSerValThrCysLeuSerLeuLeuSerValTrpThrLysLeuArgThrArgGlyArgLeuProPro   35

GGGCCCCACCCCCCTTCCCATTATTGGGAATCTACTGCAACTGAACCTCAAGGACATCCTGCATCGCTTTCCAAGCTGGCCAAAGAGTACGGGGCTGTTTACACGCTGTACTTTG  230
GlyProHisProProSerHisTyrTrpGluSerThrAlaThrGluProGlnGlyHisProAlaSerLeuSerLysLeuAlaLysGluTyrGlyProValTyrThrLeuTyrPheG   74

GAACCTCTCCTACTGTGGTCCTGCATGGCTATGATGTGGTGAAGGAGGTGCTTCTCCAGCAAGGGGATGAGTTCCTCGGGAGAGGACCTCTGCCCATCATCGAAGATACCCATAA  345
lyThrSerProThrValValLeuHisGlyTyrAspValValLysGluAlaLeuGlnGlnGlyAspGluPheLeuGlyArgGlyProLeuProIleIleGluAspThrHisLy   112

GGGATATGGACTCATTTTCAGTAATGGAGAGAGGTGGAAAGTCATGCGACGCTTCTCCCTCATGACTCTGAGGAACTTCGGAATGGGGAAGAGAAGCCTAGAGGAGAGGGTGCAG  460
sGlyTyrGlyLeuIlePheSerAsnGlyGluArgTrpLysValMetArgArgPheSerLeuMetThrLeuArgAsnPheGlyMetGlyLysArgSerLeuGluGluArgValGln   150

GAGGAAGCCTGGTGCCTTGTGGAGGAATTACAAAAAACAAAAGCTCAGCCCTTTGACCCCACCTTCATCCTGGCCTGTGCTCCCTGCAACGTGATCTGCTCCATTCTTTTCAACG  575
GluGluAlaTrpCysLeuValGluGluLeuGlnLysThrLysAlaGlnProPheAspProThrPheIleLeuAlaCysAlaProCysAsnValIleCysSerIleLeuPheAsnA  189

ATCGATTCCAGTACAACGACAAGACGTTCCTCAACGTTATGGATCTTTTAAATAAAAATTTTCAACAAGTGAACTCCGTGTGGTGCCAGATGTACAACCTTTGGCCAACGATCAT  690
spArgPheGlnTyrAsnAspLysThrPheLeuAsnLeuMetAspLeuLeuAsnLysAsnPheGlnGlnValAsnSerValTrpCysGlnMetTyrAsnLeuTrpProThrIleIl   227

AAAGTATCTCCCTGGAAAGCATATAGAATTTGCAAAAAGAATTGATGATGTTAAAAACTTCATTCTTGAAAAAGTGAAGGAACACCAGAAGTCCCTGGATCCTGCCAACCCTCGG  805
eLysTyrLeuProGlyLysHisIleGluPheAlaLysArgIleAspAspValLysAsnPheIleLeuGluLysValLysGluHisGlnLysSerLeuAspProAlaAsnProArg   265

GACTACATTGACTGTTTTCTCAGCAAGATTGAGGAGGAGAAGGACAACCTGAAGTCTGAGTTCCATCTGGAGAACTTGGCTGTCTGTGGGTCTAACTTGTTTACGGCAGGCACCG  920
AspTyrIleAspCysPheLeuSerLysIleGluGluGluLysAspAsnLeuLysSerGluPheHisLeuGluAsnLeuAlaValCysGlySerAsnLeuPheThrAlaGlyThrG   304

AGACAACCAGCACCACCCTGAGATTCGGGCTCCTGCTCCTTATGAAGTATCCAGAGGTGCAAGCCAAAGTTCATGAGGAACTTGACCGTGTGATTGGACGCCACCAACCCCCCAG 1035
luThrThrSerThrThrLeuArgPheGlyLeuLeuLeuLeuMetLysTyrProGluValGlnAlaLysValHisGluGluLeuAspArgValIleGlyArgHisGlnProProSe   342

CATGAAGGACAAGATGAAGCTGCCTTATACCGATGCTGTATTGCATGAGATTCAAAGATACATCACTCTGGTTGGTTCCAGTCTGCCCCATGCTGTGGTCCAGGACACAAAATTC 1150
rMetLysAspLysMetLysLeuProTyrThrAspAlaValLeuHisGluIleGlnArgTyrIleThrLeuValGlySerSerLeuProHisAlaValValGlnAspThrLysPhe   380

AGAGACTATGTCATCCCCAAGGGTACTACTGTACTCCCGATGCTGTCTTCCGTCATGCTGGATCAAAAGGAGTTTGCCAACCCAGAGAAGTTTGATCCAGGACACTTTCTGGATA 1265
ArgAspTyrValIleProLysGlyThrThrValLeuProMetLeuSerSerValMetLeuAspGlnLysGluPheAlaAsnProGluLysPheAspProGlyHisPheLeuAspL   419

AAAATGGCTGCTTCAAGAAGACAGACTACTTTGTTCCCTTCTCCTTGGAAAGCGGGCCTGTGTTGGTGAGAGTTGGCCCGGATGGAGCTCTTCCTGTTCTTCACCACCCTTCT 1380
ysAsnGlyCysPheLysLysThrAspTyrPheValProPheSerLeuGlyLysArgAlaCysValGlyGluSerLeuAlaArgMetGluLeuPheLeuPhePheThrThrLeuLe   457

GCAAAAGTTTTCCCTGAAGACTCTGGTGGAGCCCAAGGACCTTGACATCAAGCCTATTACTACCGGGATTATCAATTTGCCGCCACCTTACAAGCTGTGCCTTGTTCCTAGATAA 1495
uGlnLysPheSerLeuLysThrLeuValGluProLysAspLeuAspIleLysProIleThrThrGlyIleIleAsnLeuProProProTyrLysLeuCysLeuValProArg     494

GGGTTTATCCTTTTAATCATTCAAATCAAAGTAATTGTTTCCTTTTTCTATAAATATTCTTTTTATGCTAAGCACTGCCTAGGCCAGAAACATCATGCATGAAAACCACATGGAC 1610

ATTTATTTCGATTTAAAAATCTAGGTCTTAAAAGCTTCCAGGACACAGGCAGTGTGTCTGTGCCCTCAACTGCTCTAGAAACCAGGCCCCAAGCACAACTGACACCAGATATGC 1725

AAGACCCTCATCAGACGCCACCTACAAGTAGCACTGTGTGCTCTTCTGTAGCTTTGGAAATAGAATTGTTTTCTGTCATTGTTTCAATAGAGAAATTAATGTAATGGCCTACGTT 1840

CATTTTCCTGTTCACACTTAAAAACATTTACATGATTTGTGCCACATTCACAAAATAAATTCTCACCCTGACAATAAAAAAAAAGGAATTCCTGTTGGCCGAGTTGTCAGTCAGC 1955

GTGAACTCTGCCACCAGCTCCCTAAGAAGGGGCATTGAAGGACCCTTCATAGGTTTTTGGAGGTAATAAAGCCAGGATATCATAGAGCCTTAAGCGGACCATTGCAGCACTAGCT 2070

TTCAGGTGGGGGAATTCCAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAGGAATT 2153
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Figure 1. The conserved amino acids (2) of the putative heme-binding region are underlined.