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Expression of the *Bgp* gene and characterization of mouse colon biliary glycoprotein isoforms

(Carcinoembryonic antigen gene family member; reverse transcription and polymerase chain amplification; mouse hepatitis virus receptor; adhesion molecule)

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Received by: J.A. Engler: 9 October 1992; Revised/Accepted: 7 December/8 December 1992; Received at publishers: 1 February 1993

SUMMARY

The biliary glycoprotein (BGP)-encoding gene is a member of the human carcinoembryonic antigen (CEA) gene family. We have now cloned several mouse Bgp cDNAs from an outbred CD8-1 mouse colon cDNA library, as well as by reverse transcription-PCR amplification of colon RNA. The distinguishing features of the deduced Bgp protein isoforms are found in the two divergent N-terminal domains, the highly conserved internal C2-set immunoglobulin domains, and an intracytoplasmic domain of either 10 or 73 amino acids (aa). The cDNA structures suggest that these mRNAs are produced through alternative splicing of a Bgp gene and the usage of multiple transcriptional terminators. The Bgp deduced aa sequences are highly homologous to several well characterized rat hepatocyte proteins such as the cell CAM105/ecto-ATPase/pp120/HA4 proteins. Oligodeoxyribonucleotide probes representing the various cDNA isoform domains revealed predominant transcripts of 1.8, 3.1 and 4.0 kb on Northern analyses of mouse colon RNA; some of these bands are actually composed of several co-migrating transcripts. The transcripts encoding the long intracytoplasmic-tailed Bgp proteins are expressed at one-tenth the relative abundance of the shorter-tailed species. We have previously demonstrated that several mouse Bgp cDNAs, when transfected into eukaryotic cells, express BGP proteins at the cell surface and function in vitro as cell adhesion molecules, much like their human and rat counterparts. The expression of the many Bgp isoforms at the surface of epithelial cells, such as colon, suggests that these proteins play a determinant role, through self- or heterologous contact, in renewal and/or differentiation of their epithelia.

INTRODUCTION

Carcinoembryonic antigen (CEA); (Gold and Freedman, 1965) is a human tumor marker used to evaluate recurrences of gastrointestinal, breast and lung cancers (Shuster et al., 1980). The CEA gene family, composed of at least 22 genes clustered on human chromosome 19q13.1-q13.2, is divided into two subgroups

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Abbreviations: aa, amino acid(s); BGP, biliary glycoprotein; Bgp, mouse biliary glycoprotein; BgpX, gene isoforms encoding mouse Bgps (replaces mmCGM to conform with mouse genome nomenclature; X is assigned by order of characterization); bp, base pair(s); CD, complement determining; CEA, carcinoembryonic antigen; CGM, CEA-related gene family member; Cyt, intracytoplasmic tail; ICAM-1, intercellular adhesion molecule 1; Ig, immunoglobulin; kb, kilobase(s) or 1000 bp; MHV, mouse hepatitis virus; MHVR, MHV receptor; NCA, non-specific cross-reacting antigen; nt, nucleotide(s); oligo, oligodeoxyribonucleotide; ORF, open reading frame; PCR, polymerase chain reaction; PSG, pregnancy-specific glycoprotein; 5' or 3'UTR, 5' or 3' untranslated region; RT, oligo specific for Cyt in the antisense orientation; TM, transmembrane.
based on sequence comparisons and expression patterns (Thompson et al., 1992; Khan et al., 1992): the CEA sub-
group [CEA, NCA, BGP, CGM6 and a number of genes
with as yet undefined gene products (designated CGMs;
Thompson et al., 1992)] and the pregnancy-specific (PSG)
subgroup (Khan et al., 1992). The CEA-related gene pro-
ducts exhibit structural features of the immunoglobulin
(Ig) supergene family with N-terminal domains resem-
bling variable regions and internal domains homologous
to C2-set constant domains (Williams and Barclay, 1988).
BGPAs (Svenberg, 1976; Svenberg et al., 1979; Hinoda
et al., 1988; Barnett et al., 1989) are unique in this family
in that all isoforms, produced through alternative splicing
of a single gene (Barnett et al., 1989), bear either short
(10 aa) or long (71 aa) intracytoplasmic tails (Cyt)
(Hinoda et al., 1988, Barnett et al., 1989). In contrast,
CEA, NCA and CGM6 are linked to the membrane by
a glycoporpholipid anchor (Hefta et al., 1988; Kolbinger
et al., 1989; Berling et al., 1990).

CEA, NCA and BGP are postulated to participate in
intestinal tissue organization during embryonic develop-
ment and function in vitro as intercellular adhesion mole-
cules (Benchimol et al., 1989; Oikawa et al., 1989; Oikawa
et al., 1992; Rojas et al., 1990; Zhou et al., 1990). The
CEA and NCA glycoproteins are also involved in the
bacterial recognition and in colonization (Leusch et al.,
1991). PSGs are postulated to act as immunomodulators
during pregnancy (Cerni et al., 1977).

To pursue functional investigations, we have begun
characterization of the mouse CEA-related gene family.
In previous work, we have shown that mouse Bgp pro-
teins (the cDNAs had previously been called mmCGM1a
and mmCGM2 and have been renamed BgpA and BgpB),
expressed at the surface of transfected cells, function in
vitro as adhesion molecules (Turbide et al., 1991;
McCuaig et al., 1992). Moreover, Dveksler et al. (1991)
have demonstrated that one of these mouse Bgp proteins
(MHVR with identical coding sequence to the deduced
aa sequence of BgpA) acts as the mouse hepatitis virus
(MHV) receptor (Dveksler et al., 1991).

Contrary to human BGP isoforms which contain the
same Ig-like N-terminal domain (Barnett et al., 1989), the
previously reported mouse Bgp-like cDNAs each encode
a distinct N-terminal domain, and internal C2-set
domains that are conserved but not identical (Turbide
et al., 1991; McCuaig et al., 1992). This finding implied
that either a Bgp gene subfamily exists, or that complex
transcriptional mechanisms are operative to account for
these Bgp cDNAs. It therefore became important to
define the number of Bgp isoforms and to study their
expression in mouse tissues for future functional studies.
The data presented in this paper describes the character-
ization, the relative abundance and the differential expres-
sion of nine Bgp cDNA isoforms expressed in colon. The
results suggest that a unique mouse Bgp gene undergoes
alternative splicing, and that different polyadenylation
signals are used to produce a number of related proteins
with possible complementary functions. However, the
two classes of related Bgp cDNAs cloned from the out-
bred CD-1 mouse may represent allelic variants of this
Bgp gene (Dveksler et al., 1993).

RESULTS AND DISCUSSION

(a) Library cDNA clones

To characterize the mouse CEA-related gene family,
we screened a CD-1 mouse colon cDNA library
(Beauchemin et al., 1989). This yielded a significant
number of cDNA clones (i.e., 392), some of which pre-
sented different restriction patterns or different cDNA
sequences. To verify if these cDNA clones were represen-
tative of other mouse CEA-related genes, clones 23, 32,
37, 58 and 64 were completely sequenced while clone 132
was partially sequenced. As shown in Fig. 1, some of these
clones (clones 23 and 132) overlapped perfectly within
sequences encoding either the signal sequence, the N ter-
mainal or the A2 domain of the BgpA cDNA
(formerly mmCGM1a) (McCuaig et al., 1992), while
others (clones 23, 32, 37, 58 and 64) were aligned with
dNA coding for either the leader, the N terminal, the A2,
the TM, the Cyt domains or the 3'UTR sequence of
another published cDNA clone, BgpB (formerly
mmCGM2) (Turbide et al., 1991). Clone 23 extended the
published BgpB cDNA 3'UTR (Turbide et al., 1991) by
119 nt and ended in a poly(A) tail. Another cDNA clone
(clone 58) encoded a longer Cyt (73 aa) than the pre-
viously reported 10 aa Cyt domain (Turbide et al., 1991;
McCuaig et al., 1992); this new mouse Cyt domain, how-
ever, resembled closely the cDNA sequence of human
BGP (Barnett et al., 1989) and of a rat ecto-ATPase cDNA
(Lin and Guidotti, 1989). Clones 32 and 64 also encoded
conserved but not identical A1 and B1 domains when
compared to similar domains of the BgpA cDNA. cDNA
clone 37 demonstrated high homology to the reported 3'UTR
of the MHVR isolated from Balb/c mice (Dveksler
et al., 1991), but extended this sequence by 199 nt to
another poly(A) tail.

(b) RT-PCR amplification

The information from the cDNA sequences and com-
puter-translated aa sequences compiled from the cDNA
clones were indicative of multiple Bgp cDNA isoforms
present in mouse colon. We therefore conducted a series
of experiments using a RT-PCR amplification technology
(Frohman et al., 1988) to identify the type of Bgp isoforms
Methods: cDNA was synthesized as previously described (Beauchemin et al., 1989). The CD-1 mouse colon cDNA library was screened with cDNA restriction fragments corresponding to the EcoRI-SstI and Accl-EcoRI restriction fragment of clone 46 (McCuaig et al., 1992) and with the oligo RIT (antisense: S-TGAGGGTTTGTGCTCTGTGAGATC) representing a region of the long Cyt conserved between human BGP (Barnett et al., 1989) and rat ecto-ATPase (Lin and Guidotti, 1989). Bgp cDNA restriction fragments were subcloned into unique sites of the BluescriptSK+ plasmid (Stratagene, La Jolla, CA) and overlapping DNA restriction fragments were sequenced on both strands with either the T7 or the T3 promoter primers or with internal primers by the dideoxy chain termination method (Sanger et al., 1977) using T7 DNA polymerase (Pharmacia, Montreal, Canada). Sequences were analyzed using the DNAsis, Prosis (Pharmacia) and the GCG sequence analysis system (Devereux et al., 1984) programs.

present in this tissue. Total RNA from CD-1 mouse colon was reverse-transcribed using either a (dT)$_{17}$ adaptor primer or KM5, a primer located in the 3' UTR of BgpB and BgpD (see Fig. 3b). PCR amplifications were then performed on single-stranded cDNAs with combinations of primers located in the coding and non-coding regions and the products were cloned and sequenced (Fig. 2a). Since we had observed that major sequence differences were found between the N-terminal domains (designated N1 and N2; Turbide et al., 1991; McCuaig et al., 1992), divergent primers within these two N-terminal domains (46N1, R46N1, CGM2N and RCGM2N) were synthesized, tested at medium and high hybridization stringency and demonstrated non-cross-reactivity (data not shown). The coding sequence of the two A1 domains (A1a, A1b), B1 domains (B1a, B1b) and the two A2 domains (A2a, A2b) differed by only 4, 1 and 3 nt, respectively, (indicated in Fig. 3b). Common primers specific for each of these domains (KM7 and KM8 for A1 and 33-35 for A2) as well as a primer specific to the region encoding the long Cyt (RIT) were synthesized.

The use of primers specific to the 5' UTR of BgpA (KM2), and to the N-terminal domains (46N1 or RCGM2N) resulted in the amplification of a 360-bp fragment only when KM2 was combined with a primer from the N1-terminal domain (46N1). This result indicates that the 5' UTR of the transcripts containing the N2 domain is different in the region of KM2 (Fig. 2a). Varying the Mg$^{2+}$ concentrations of the amplification reaction for the KM2-RCGM2N primer pair did not produce a fragment. The identity of the KM2-46N1 fragment was confirmed by sequencing of the fragment.

The sequences of the cDNA clones suggested that splicing events occurred on the primary Bgp transcript(s), resulting in the deletion of the A1 and B1 domains (McCuaig et al., 1992), or in the inclusion of an exon to generate a long Cyt protein. To ascertain the numerous splicing possibilities downstream from the N-terminal
domains, amplifications were carried out between N-terminal primers (R46N1 or CGM2N) and oligos corresponding to the A2 domain (33–35), the long Cyt (RIT) or one of the 3'UTR's (KM6). As is summarized in Fig. 2a, amplification between primers R46N1 and 33–35 or CGM2N and 33–35 resulted in the synthesis of two different-sized fragments in each case. Cloning and sequencing revealed that the shorter-sized fragments were derived from cDNAs where the N-terminal coding domains, N1 or N2, were followed by their respective A2 domains (N1 domain with A2a, N2 domain with A2b). None of the sequenced clones spliced either the N1 domain to the A2b domain, or the N2 domain to the A2a domain. Sequencing of the longer fragments cloned from each amplification demonstrated similar specificity; the N1 domain only associated with the A1a and B1a domains followed by the A2a domain while the N2 domain was joined to the A1b, B1b and A2b domains.

Similarly, amplifications using the N terminal domain and the long Cyt-specific primers produced two DNA fragments. Sequencing confirmed that the shortest ones carried a deletion of the A1 and B1 domains, while the longer fragments included these two domains. Amplifications conducted with primers located in the N-terminal coding domains and in the 3'UTR (KM6) confirmed the previous results. A greater number of clones (seven) representing these fragments were sequenced, one of which was shown to code for a long Cyt domain. Results from these amplifications were confirmed by Southern analyses. As can be seen in Fig. 2b,
the 1350 and 1450-bp fragments amplified by either pairs (R46N1-KM6 or CGM2N-KM6) hybridized to an oligo specific to the A1 domains (KM7) (Fig. 2b, panel A), while all the fragments hybridized to the 33-35 oligo defining the A2 domain (Fig. 2b, panel B). The bands hybridizing to oligo RTT (Fig. 2b, panel C) confirmed that the short-tailed cDNA isoforms have a counterpart encoding a long Cyt. These hybridizations also revealed a new 3.0-kb DNA fragment specifically amplified in primer combinations containing the N1 primer. This novel cDNA fragment will be the subject of future reports. This fragment is not due to contamination with genomic DNA since it was not present in PCR amplifications when the RT reaction was omitted (data not shown).

(c) The nt sequences of the cDNAs and the aa sequences

The structure of the deduced isoforms are depicted in Fig. 3a. A minimum of nine cDNA isoforms are generated from the mouse Bgp gene(s). Two of these cDNA isoforms (BgpA and MHVR) have an identical coding region but somewhat different 5' and 3'UTRs (the first 41 nt upstream from the ATG start codon are identical) (Dveksler et al., 1991; McCuaig et al., 1992). As was demonstrated by Southern analysis (Fig. 2b, panel C) and sequencing of the PCR products, cDNAs which lack a region encoding two C2-set domains such as BgpB and BgpC (clones 1 and 23 respectively) can also encode a long Cyt to generate the BgpG and BgpH cDNA isoforms. Likewise, the Southern and sequencing analyses indicate that the regions encoding the long Cyt as well as three internal repeats are present in the two longest cDNA isoforms designated BgpD and BgpF (Fig. 3b).

The Bgp isoforms exhibit two variants of the signal sequence: four-nt substitutions (Fig. 3b) lead to change in two aa (Fig. 3c). The leader region is followed by an N-terminal domain. There are two possible N-terminal domains (N1 and N2) which exhibit both common and divergent areas: the nt sequences encoding the first 37 aa of the N-terminal domain exon is paired with the first two nt downstream, cDNA library clones terminate at threeous poly(A) tails, only one of which fits the perfect consensus sequence (AATAAA) (Proudfoot and Brownlee, 1976).

The predicted aa sequence of the Bgp isoforms are presented in Fig. 3c and may be sorted into two classes, defined by their N-terminal domains. The first 37 aa of these domains are identical; however, in the following stretch of 19 aa, nine residues are not conserved; charged structures replace non-polar aa and vice versa (K→A, M→K, F→Q). The Cys residues of the C2-set domains, thought to be involved in intrachain disulfide bonding (Williams and Barclay, 1988), are well conserved in all isoforms (shading in Fig. 3c).
Fig. 3. Topology and cDNA sequences of mouse Bgp isoforms and deduced aa sequences. (a) Topology of Bgp isoforms. Abbreviations used have been described in legend to Fig. 1. MHVR, mouse hepatitis virus receptor. (b) cDNA sequences. The nt sequences of the longest isoforms (BgpD and BgpF) are presented. Dots indicate identical nt. The sequences of the oligos used as primers or probes are indicated by lines above or below the nt sequences and their orientation by arrowheads on these lines. The ATG start and TGA stop codons are boxed. The domains are separated by lines and captions over and under the sequences. Open arrowheads delineate the 53 bp exon inserted to generate the long Cyt domain. The nt 1534–1550 are common to 3'UTR1 and 3'UTR2. The asterisk (at nt 1550) represents the position where the 3'UTR1 (McCuaig et al., 1992) replaces the 3'UTR2. Double underlines indicate putative or actual poly(A) consensus signals. The poly(A) tail of cDNA clone 23. The poly(A) tail at nt 2908 is that of cDNA clone 37. Sequence differences between nt 1747–1829 in the 3' UTR between various clones are indicated at the bottom of the figure. GenBank accession Nos.: BgpC, X67278; BgpD, X67279; BgpE, X67280; BgpF, X67281; BgpG, X67282; BgpH, X67283.

(c) Deduced aa sequences of Bgp. The nt sequences were computer-translation and the deduced aa sequences were aligned. Double arrowheads indicate delineation of the domains. The protein sequences are written in the one letter aa code. aa positions are indicated above the signal sequences and N-terminal domains. Glycosylation sites are underlined. Shaded C represent the conserved Cys residues. Arrow indicates position of the nt modification leading to a silent mutation.

Methods: The full coding cDNAs for BgpD, BgpE, BgpF, BgpG, BgpH were reconstructed by ligating overlapping cDNA clones at various restriction sites. Reconstructed cDNA clones were completely resequenced to ensure accuracy of sequence.
Fig. 4. Northern analyses and quantification. (a) Positions of oligos in the BgpD and BgpF isoforms. The positions of the oligos in the various regions of the cDNAs are indicated by arrows below the structures. (b) Northern analyses. Separation of mouse colon total RNA on a formaldehyde-agarose gel and hybridization with oligos. Positions of the 18S, 1.86-kb and 28S 4.71 kb as well as the three major transcripts quantitated in c are indicated. (c) Quantification. Autoradiograms of different exposure times of the above Northern blots were scanned by laser densitometry using a Bio-Imager scanner (Millipore, Montreal, Canada). The 4.0-kb band revealed by hybridization to oligo 46N1 was weighed as 1.0. All other readings were computed proportionally. n.d., not determined. Methods: 20 μg of total colon RNA, prepared by guanidium isothiocyanate extraction and centrifugation, was electrophoresed through a 2.2 M formaldehyde-1.5% agarose gel and transferred to GeneScreen Plus membranes. Hybridizations were carried out as previously described (McCuaig et al., 1992) with the [32P]oligos (Sambrook et al., 1989) indicated above the autoradiograms. The filters were washed to a final stringency corresponding to 5°C below the predicted melting temperature of each oligo.

(d) Northern analysis and relative abundance of transcript isoforms

Northern analyses were performed on mouse colon transcripts with oligos representing different domains of the cDNAs (Fig. 4a and b) to quantify the relative abundance of the Bgp isoforms (Fig. 4c). Three major transcripts were identified with these probes. These were measured as 1.8, 3.1 and 4.0-kb relative to rRNA markers. In comparison, human BGP restriction fragments have been shown to hybridize to 2.2 and 3.9-kb transcripts from a variety of cell lines (Hinoda et al., 1988; Barnett et al., 1989). The nt sequences of BgpD and BgpF isoforms reported in Fig. 3b are 2922 nt long, while the BgpB and BgpC or BgpG and BgpH isoforms are 1518 or 1569 nt long, respectively. The size discrepancy between the cDNA sequences and the transcript lengths measured by Northern analysis may be due to an additional 300 nt at the 5' end (P.N. and N.B., unpublished results). As well, the length of the poly(A) tails are thought to be 400-bp longer than the DNA sequences of the human CEA gene family members (Beauchemin et al., 1987). This adjustment would be consistent with a size of 3622-bp for the longer cDNAs. The existence of an even longer 3'UTR has not been ruled out. The 4.0-kb transcripts and a strong hybridization signal at 3.1-kb with a relative abundance of 1.5 are detected with oligo KM6; the 3.1-kb length is compatible with cDNAs expressing L-N-A2-TM-CytS or CytL carrying a complete 3'UTR region.

To quantify the number of transcripts co-migrating in the revealed bands, equal pmol of the labelled oligos were added to hybridization mixtures to ensure that the intensity of the bands would reflect the relative abundance of the transcripts. The most striking observation was that, although the cDNAs encoding the isoforms containing only two Ig domains (N and A2) could be readily cloned from a cDNA library or amplified by PCR, their abundance as 1.8-kb transcripts was minimal (Fig. 4c: 46N1, RCGMN2 and KM7). The N1 and N2-terminal domains are preferentially expressed with three Ig C2-set constant domains as full-length transcripts. But, the transcripts encoding proteins with long Cyt are preferentially expressed without A1 and B1 domains and migrate as 1.8-kb rather than 4.0-kb mRNA. These are 1/10 to 1/3 as abundant as the short Cyt-encoding isoforms. As expected, hybridization with an oligo (3UTR-2) located in the distal region of the 3'UTR detects preferentially 4.0 kb transcripts.

An oligo specific for the A2 domain (33-35) binds to both the 1.8 and 4.0-kb transcripts; however, quantification of the autoradiograms at different exposure times indicated exceedingly high values for this domain. Although the migration of these transcripts is similar to the 18S rRNA, the signal seen does not represent cross-hybridization to rRNA; when this oligo was used to probe non Bgp-expressing cell lines, no signal was encountered. However, this A2 domain is very conserved when compared to a similar domain expressed in the mouse Psg cDNAs (Kodelja et al., 1989; Rebstock et al., 1990; Rudert et al., 1992). Thus, the oligo 33–35 may be detecting some of these PSG-encoding transcripts.
(e) Conclusions

1. The data presented in this report emphasize the multiplicity of the mouse Bgp isoforms and suggest mechanisms responsible for their generation. So far, nine similar, but not identical cDNA isoforms have been cloned from CD-1 mouse colon (Turbide et al., 1991; McCuaig et al., 1992; this report) and from Balb/c liver (Dveksler et al., 1991). Alternative splicing is active in the generation of isoforms exhibiting different Cyt domains as well as isoforms lacking internal A1 and B1 domains. Northern analysis performed on mouse colon RNA with specific oligo probes and quantification of the autoradiograms have confirmed the diversity of the Bgp isoforms; three major transcripts (1.8, 3.1 and 4.0 kb) encode these isoforms, albeit in different relative amounts. The relative abundance of the Bgp long Cyt-containing transcripts are approximately tenfold less abundant than the short Cyt-tailed isoforms. This finding may reflect on the function of the long-tailed BGP proteins since they contain consensus Tyr phosphorylation sites that have been conserved throughout evolution and are present in the mouse, the rat and the human BGP counterparts (Lin et al., 1989; Afar et al., 1992; Culic et al., 1992). Overexpression of these isoforms may be detrimental to cell growth and/or differentiation.

2. In contrast to what has been described for human BGP isoforms (Barnett et al., 1989) there are, in fact, two classes of mouse Bgp isoforms. Sequence analyses performed on the cDNA isoforms from CD-1 mice have demonstrated that the N1-terminal domain is only associated with either the A1a or A2a domain, while the N2-terminal domain is joined to the A1b or A2b domain. These C2-set Ig domains differ from each other by only a few nt while the N-terminal domains exhibit a greater number of changes. The A2 domain is also a required structural feature of the mouse colon Bgp isoforms contrary to the human BGPb and BGPld isoforms (Barnett et al., 1989) which lack this domain as do BGPg, BGPh, BGPx and BGPy (T. Barnett, personal communication). However, all isoforms examined in this report carry identical TM and long or short Cyt domains. The length of the 3' UTR varies but the sequence within remains invariable (except for a few nt modifications in a highly repetitive region), strongly suggesting that a single Bgp gene encompasses all exons.

However, the generation of isoforms bearing a few nt modifications could also be explained by allelic variation. The present characterization was done on the outbred CD-1 mouse. A recent report suggests that expression of the two classes of isoforms (defined by their N-terminal domains) are due to allelic variation of the same gene. Balb/c mice do not express the N2-containing cDNA isoforms and conversely, SJL/J mice lack the N1-bearing isoform transcripts and proteins (Dveksler et al., 1993). The splicing versus allelic variation issue will be resolved with the characterization of the complete Balb/c Bgp gene (P.N. and N.B., in progress).

3. The feature which renders the two isoform classes distinct lies in their N-terminal domains. The first 37 aa of these domains are identical, while the last third encloses many modifications mostly concentrated in a central core. Recently, Bates et al. have constructed a three-dimensional model for human CEA based on the NMR structure of the rat CD2 as well as X-ray crystal structures of human CD4 and Ig variable domains (Bates et al., 1992). The first two domains of CEA are predicted to have a rod-like appearance with numerous tightly packed β sheets and a few exposed loops on either side of the molecule. Interestingly, the most divergent aa residues (positions 72–90) within the mouse Bgp N-terminal domains would be localized in the C-C' loop which is predicted to be exposed. On the other hand, some of the conserved 37 aa are positioned within another exposed loop. These loops are the most likely targets for interactions with other cells or ligands. In fact, two described properties of these mouse glycoproteins are dependent upon such interactions; two protein products of the Bgp cDNA isoforms (BgpA and BgpB) can function in vitro as cell adhesion molecules (Turbide et al., 1991; McCuaig et al., 1992) and one (MHVR or BgpA) has also been shown to bind to MHV spike glycoprotein (Williams et al., 1990; Dveksler et al., 1991). Although other domains of the CEA/BGP proteins are necessary for intercellular adhesion, the N-terminal domain appears to be critical for this function (Oikawa et al., 1991). Similarly, other members of the Ig supergene family (CD4, ICAM-1, CD2, poliovirus receptor) require that their N-terminal domains enter into contact with either viral envelope glycoproteins (Greve et al., 1989; Mendelsohn et al., 1989; Staunton et al., 1989; White and Littman, 1989; Koike et al., 1990; Pollard et al., 1991) or their respective ligands to be functionally activated (Staunton et al., 1990; Koike et al., 1991; Regiser et al., 1991).

Human BGP mRNAs and proteins appear to be less abundant than CEA in colon but more abundant in liver (Hinoda et al., 1990; Drzeniek et al., 1991). Extensive expression analyses of these two protein entities have been hampered by the lack of specific anti-BGP antibodies which have only recently been developed (Drzeniek et al., 1991). However, the mouse Bgp mRNAs (Turbide et al., 1991; McCuaig et al., 1992) and proteins (M.R., P.N., S.J., C.T., D.F. and N.B., manuscript in preparation) are more abundant in colon than liver. Since a true CEA homolog or any of the phosphatidylinositol-linked CEA family members have yet to be identified in the mouse or rat models, it is tempting to speculate that the functions...
associated with CEA in the human may represent a recent evolutionary event (Stanners et al., 1992) and that these functions may be ensured in the mouse by other Bgp-related genes (P.N. and N.B., in preparation). The significance of the concurrent expression of the many Bgp isoforms on the surface of mouse tissues such as colon, intestine, liver and uterus (Turbide et al., 1991; McCuaig et al., 1992) may be that these proteins play a determinant role, through self- or heterologous contact, in renewal and/or differentiation of their epithelia.

ACKNOWLEDGEMENTS

The authors wish to thank Dr. Annette Herscovics and Dr. Clifford Stanners for critical reading of the manuscript and Dr. André Veillette for the gift of the NIH3T3 cells. K. McC. is a recipient of a studentship from the Medical Research Council of Canada. M.R. from the 'Institut Méricus', Lyon, France, N.B. is a scholar of the 'Fonds de la Recherche en Santé du Québec'. This research is supported by the Medical Research Council of Canada and The Cancer Research Society Inc.

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