

SHORT COMMUNICATION

Assignment of the Protein Kinase C δ Polypeptide Gene (PRKCD) to Human Chromosome 3 and Mouse Chromosome 14

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The protein kinase C (pkc) enzymes are a family of serine-threonine protein kinases, each encoded by a distinct and separate gene. The chromosomal locations of human *PRKCA*, *PRKCB*, and *PRKCG* have previously been established. We now report that *PRKCD*, a novel member of the pkc gene family, maps to human chromosome 3. The chromosomal location of *Pkcd* has also been determined in the mouse by analysis of recombination frequency in an interspecific panel of backcross mice. We find that the locus encoding *pkcd* resides proximal to nucleoside phosphorylase (*Np-2*) and *Tcra* on mouse chromosome 14 in a region syntenic with human 3p. © 1994 Academic Press, Inc.

The pkc genes can be subdivided into two major classes, conventional (α , $\beta 1$, $\beta 2$, and γ) and novel (δ , ϵ , ζ , η , and θ), based on their sequence and biochemical properties (1-4). Although the sequence homology among the pkc family of genes is extensive, the pattern of expression varies among tissues. For example, *pkcd* appears to be the major isoform expressed in mouse hematopoietic cells (5). Thus, the pkc family of genes must fall under the constraints of a complex system of regulation. It has already been reported that conventional pkc genes map to human chromosomes 17q [*PRKCA* (6, 7)], 16p [*PRKCB1* (6)], and 19q [*PRKCG* (6, 8)], respectively. As part of our interest in pkc expression in hematopoietic cells, we have recently isolated and characterized the gene encoding *pkcd* (5). The *pkcd* protein is activated by phospholipid, yet does not require Ca^{2+} , apparently as a result of the absence of a C2 domain. To characterize further the *PRKCD* gene, we have identified and compared the chromosomal position of *PRKCD* to those of the other conventional isoforms of pkc.

Originally, a 2.5-kb cDNA clone specific to mouse *pkcd* was isolated from the myeloid tumor ABPL2 with a 0.38-kb PCR-generated subclone of *pkcd* (5). This insert was determined to contain nearly a full-length copy of

the mouse *pkcd* mRNA and does not cross-hybridize to other pkc genes under stringent conditions. To identify the human locus (*PRKCD*) corresponding to *pkcd*, we hybridized the mouse cDNA probe to a panel of somatic cell human-hamster hybrid DNAs. The *pkcd* probe hybridizes to a single 23-kb *Bam*HI band in hamster DNA and two *Bam*HI fragments of 8.5 and 3.0 kb in human DNA. Comparison of these bands to *Bam*HI fragments generated by pkc probes from other isoforms reveals no overlapping bands under similar stringencies, indicating that these hybridizing fragments are specific to *pkcd*. Further examination of the panel of somatic cell hybrids reveals that two hybrids, namely, 507 and 1079, retain human chromosomes that contain *PRKCD* (Table 1). The only chromosome consistent with this observation is human chromosome 3.

We utilized an interspecific backcross panel of mice developed in our laboratory to determine the chromosomal location in the mouse. The *pkcd* probe hybridizes to a single *Eco*RI fragment of 5.0 kb, indicating that the entire mouse *pkcd* gene is probably found within this segment in the genome. We were also able to identify specific restriction fragment length polymorphisms (RFLPs) between the parental mouse strains [BALB/cAn and *Mus spretus* (Spain)] with the restriction endonucleases *Xba*I, *Taq*I, and *Pvu*II for *pkcd*. We utilized a *Taq*I RFLP to follow the segregation patterns in a panel of backcross individuals. The segregation pattern generated by the *pkcd* RFLP indicates that the gene encoding *pkcd*, *Pkcd*, is most likely found on mouse chromosome 14. Analysis of recombination frequencies and reduction of double crossover events indicate that *Pkcd* maps 10.9 ± 3.6 cM (8/73 recombinants) proximal to the nucleoside phosphorylase (*Np-2*) locus and approximately 12.3 ± 5.0 cM proximal to the *Tcra* locus (Table 2). Interestingly, localization of *Pkcd* to the proximal end of mouse chromosome 14 falls within a region of known synteny with human chromosome 3p (9). Thus, we predict that *PRKCD* most likely maps to the short arm of human chromosome 3.

In conclusion, we find that the chromosomal location of pkc genes appears to be dispersed in both human and mouse. Therefore, pkc genes must exhibit a *cis*-acting form of tissue-specific regulated expression.

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TABLE 1

Correlation between Specific Human Chromosomes and the *PRKCD* Locus in 11 Somatic Cell Hybrids

Chromosome	212	507	683	734	756	862	909	937	1006	1049	1079	<i>PRKCD</i>
1	-	-	-	-	-	-	-	+	-	-	-	-
2	-	-	-	-	-	-	-	-	-	-	-	-
3	-	+	-	-	-	-	-	-	-	-	+	+
4	-	-	-	-	-	-	-	-	55%	-	-	-
5	d	+	+	+	d	+	d	+	+	+	+	-
6	-	-	-	-	65%	-	+	-	-	-	-	-
7	-	-	-	-	+	-	-	-	+	-	-	-
8	-	-	-	-	-	-	+	-	+	-	-	-
9	-	-	-	+	-	+	-	-	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-
11	-	-	-	-	-	-	-	-	-	+	-	-
12	-	+	45%	-	+	-	-	-	-	-	-	-
13	-	-	-	-	65%	-	-	-	+	-	-	-
14	-	65%	+	-	45%	-	+	+	-	-	-	-
15	-	-	-	-	-	-	-	+	+	-	10%	-
16	-	-	-	-	-	-	-	-	-	-	+	-
17	-	-	-	-	-	-	-	+	-	-	-	-
18	-	-	-	+	-	-	-	-	-	-	-	-
19	-	-	+	-	+	-	-	-	+	-	-	-
20	-	40%	-	-	+	-	-	-	-	-	-	-
21	-	-	+	-	+	-	-	+	+	-	-	-
22	-	25%	+	-	-	-	-	-	-	-	-	-
X	-	-	-	-	-	-	+	-	-	-	-	-
Y	+	+	-	-	+	-	-	-	-	-	-	-

Note. d, portions of chromosome deleted; %, cell population percentages.

TABLE 2

Linkage Map of Mouse Chromosome 14

Marker	BALB/c					<i>Mus spretus</i>	Enzyme	
<i>Pkcd</i>	□	■	□	■	■	1.25	1.75	<i>TaqI</i>
<i>Np-2</i>	□	■	■	□	■	10.0	5.5	<i>EcoRI</i>
<i>Tcra</i>	□	■	■	□	□	1.8	2.3, 3.0	<i>HindIII</i>
	31	33	1	7	1			
	<i>Pkcd</i> -10.9 ± 3.6-							
	<i>Np-2</i> -1.4 ± 1.4- <i>Tcra</i>							

Note. BALB/cAn (□); BALB/cAn/*M. spretus* (■).

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