Based on pharmacological properties, the P2X receptor family can be subdivided into those homo-oligomers that are sensitive to the ATP analog αβ-methylene ATP (P2X1 and P2X3) and those that are not (P2X2, P2X4, P2X5, P2X6, and P2X7). We exploited this dichotomy through the construction of chimeric receptors and site-directed mutagenesis in order to identify domains responsible for these differences in the abilities of extracellular agonists to gate P2X receptors. Replacement of the extracellular domain of the αβmeATP-sensitive rat P2X2 subunit with that of the αβmeATP-insensitive rat P2X3 subunit resulted in a receptor that was still αβmeATP-sensitive, suggesting a non-extracellular domain was responsible for the differential gating of P2X receptors by various agonists. Replacement of the first transmembrane domain of the rat P2X2 subunit with one from an αβmeATP-sensitive subunit (either rat P2X2, or P2X3 subunit) converted the resulting chimera to αβmeATP sensitivity. This conversion did not occur when the first transmembrane domain came from a non-αβmeATP-sensitive subunit. Site-directed mutagenesis indicated that the C-terminal portion of the first transmembrane domain was important in determining the agonist selectivity of channel gating for these chimeras. These results suggest that the first transmembrane domain plays an important role in the agonist operation of the P2X receptor.

P2X receptors are ligand-gated ion channels activated by extracellular ATP. Although these receptors were first described almost 20 years ago (1), the lack of useful pharmacological tools has greatly hampered the elucidation of the roles that these receptors play in ongoing physiological functions (2, 3). Recent advances in the molecular biology of these receptors have led to a resurgence of interest, and have served to illustrate how little is actually known about these proteins. Indeed, results from in situ hybridization and immunochromatographic studies demonstrate that these subunits have a widespread distribution throughout the body, being present in almost all tissues (for a review, see Ref. 4), suggesting that they may have more extensive functions than appreciated previously.

To date, a total of seven individual subunit genes have been cloned and their products characterized (4). These subunits have been demonstrated to form homo- and/or hetero-oligomeric receptors (5) that are non-selective cation channels with a high permeability to Ca2+ (6), a property that confers the potential for important functions in excitable and secretory cells. Recent investigation into the structural features of the subunits has helped elucidate the role of the transmembrane domains in ion permeation through the channel (7–9), and have identified a number of extracellular residues as affecting the binding of agonists and antagonists (10–13). Nevertheless, the domains involved in ligand-induced opening of the channel (gating) have not yet been delineated, nor have the domains responsible for the pharmacological fingerprints of the various homo- and hetero-oligomeric P2X receptors.

Originally, the P2X receptor family was distinguished from other ATP receptors (such as the G-protein-coupled P2Y receptors) by their sensitivity (EC50 ≤ 30 μM) to the ATP analog αβ-methylene-ATP (αβmeATP) (14). Characterization of the cloned subunits has now shown that only two of the seven mammalian gene products have appreciable sensitivity to this analog (3). For this study, we sought to use chimeric P2X proteins to exploit the pharmacological differences between the αβmeATP-sensitive and -insensitive subunits in an effort to identify the domains and/or amino acid residues involved in determining agonist potencies at P2X subunits. Topological studies have shown that a P2X subunit has two transmembrane domains, with the intervening loop located extracellularly and the N and C termini located inside the cell (15). Previous studies from other groups have provided evidence that residues in the extracellular loop near the first transmembrane domain (TMD1) can affect the binding of ATP to the receptor (12, 13), and results from our laboratories (16) suggested that mutations in TMD1 can influence channel gating and agonist potency. In this report we present further evidence that TMD1 determines the potencies of the agonists ATP and αβmeATP in gating the P2X2 receptor, leading to the interpretation that this domain is involved in the transduction pathway linking agonist binding to opening of the channel gate in this receptor family.

MATERIALS AND METHODS

DNA Constructs—Several different types of mutant receptors were engineered, all of them through the use of overlap polymerase chain reaction as described previously (5). Briefly, chimeras were constructed by performing two successive polymerase chain reaction amplifications using 5′ and 3′ primers containing overlapping regions between rP2X2 and the specified P2X2 subunit (rP2Xr, rP2X3, or zP2X3). To generate a chimera consisting of rP2X2 with the extracellular domain replaced.
with that of rP2X3 we used oligonucleotides encoding an VYEK/SYQD
cognate sequence from one of the following: rP2X1, rP2X3, or zP2X3.

Electrophysiological Recordings—Whole-cell currents were recorded
at room temperature using the perforated-patch method. The pipette
solution contained (in mM): 130 cesium methanesulfonate, 24 CsCl, 1
MgCl2, 1 CaCl2, 10 HEPES, and amphotericin B (200 µg/ml) (17). The
extracellular solution contained (in mM): 130 cesium methanesulfonate,
24 CsCl, 1 MgCl2, 10 HEPES, and 10 glucose, and 10 HEPES. Cells predicted
to express P2X receptor protein were identified using fluorescence
microscopy to detect the presence of eGFP. Different concentrations of
ATP were applied by positioning the cell in front of one of a number of
inlet tubes arranged side-by-side in the bath chamber as described
previously (18). Successful applications of ATP were separated by at
least 3 min to minimize receptor desensitization. Concentration-response
curves were generated from the nonlinear, least squares fit to the Hill
equation as implemented in version 4.0 of Igor Pro (www.wavemetrics.com)
using the averaged data computed for each concentration of ATP. All
values given are means ± standard deviations.

RESULTS

The potency of ATP at the homo-oligomeric rP2X receptors falls
into three categories: high (EC50 = 1–3 µM, seen at rP2X1 and
rP2X3), intermediate (EC50 = 10–30 µM, seen at rP2X2,
rP2X4, and rP2X5), and low (EC50 = 100 µM, seen at
rP2X6) (3). ATP has no action at P2X6, as this subunit does not appear
to form homo-oligomeric receptors (5, 19). In contrast, the ATP
analog αβmeATP exhibits high potency and efficacy at only two
subunits (rP2X2, and rP2X4). EC50 = 1 µM), while having little or
no potency at the other five subunits even at concentrations
approaching the millimolar level (3). There are two simple
hypotheses that could explain the inability of αβmeATP to
operate five of the seven subunits: first, that αβmeATP does
indeed bind with appreciable affinity to all receptors but is
unable to induce the channel to gate in five of them (i.e. it is
functionally equivalent to an antagonist), and second, that
αβmeATP does not interact at the agonist binding domain
with appreciable affinity and thus its binding energy is not
sufficient to cause the degree of tertiary/quaternary structure
perturbation necessary to result in the gating of the channel. It was
important to determine which of the two hypotheses explains
the αβmeATP data so that experiments to investigate the
structural basis for the pharmacological differences among
subunits could be designed in a rational fashion.

The first hypothesis, that αβmeATP is a functional antago-
nist, was the most straightforward question to test, and we did
so by examining the ability of a relatively high (100 μM) concentration of αβmeATP to reduce the efficacy of a co-applied dose of 10 μM ATP at the homomeric rP2X2 receptors, at which ATP has moderate and αβmeATP little potency (Fig. 1a, ATP EC$_{50}$ = 22 μM; αβmeATP EC$_{50}$ > 300 μM). As shown in Fig. 1b, co-application of αβmeATP did not alter the degree to which a sub-EC$_{50}$ concentration of ATP activated the receptor. This result rules out the hypothesis that αβmeATP binds to the agonist site on the rP2X2 receptor with appreciable affinity. This leaves the second hypothesis, that αβmeATP’s inability to gate the rP2X2 subunit is due to a low binding affinity for the agonist binding domain, to be tested. This low αβmeATP affinity would presumably result from differences in the three-dimensional structure of the P2X2 receptor compared with the P2X1 or P2X3 receptors that arise from the evolutionarily derived amino acid variations present in their predicted primary structures. We therefore exploited this sequence divergence in testing the low affinity hypothesis by constructing chimeric receptors derived from αβmeATP-sensitive and -insensitive subunits.

**Chimeras Formed from P2X$_1$ and P2X$_2$ Subunits Show Altered αβmeATP Sensitivity**—The first chimeric receptor constructed was composed of the αβmeATP-sensitive rP2X$_2$ subunit and the αβmeATP-insensitive rP2X$_1$ subunit. This initial chimera, labeled 1.2.1, was a rP2X$_2$ subunit chain in which only the extracellular domain (presumptive site of the ATP-binding domain) was replaced with the equivalent sequence of the rP2X$_1$ subunit (schematic shown in Fig. 2a). It was anticipated that this “domain swap” would yield a homo-oligomeric receptor with the pharmacology of the P2X$_2$ receptor. In contrast to these expectations, the receptor formed from this chimeric subunit exhibited increases in both ATP and αβmeATP sensitivity (ATP: EC$_{50}$ = 0.93 μM, αβmeATP: EC$_{50}$ = 49 μM) (Fig. 2b). This unexpected result demonstrated that the extracellular domain of rP2X2 was indeed capable of forming a binding pocket at which αβmeATP could bind with both high affinity and efficacy and, by extension, that non-extracellular regions of a P2X subunit protein can profoundly influence the agonist binding and/or gating properties of the receptor.

Due to the nature of the 1.2.1 chimera, the increased agonist sensitivities observed would have to be attributable to one or more of the amino acid residue(s) present in the rP2X$_1$ portion of the molecule. There are four distinct domains of rP2X$_1$ in 1.2.1: the cytosolic N terminus, TMD1, TMD2, and the cytosolic C terminus. Recent work from our laboratories has found that point mutations in the TMD1 of rP2X$_2$ can change the potency of ATP at the receptor as well as influence the gating properties of the channel (16). In addition, data from two other groups (12, 13) strongly suggest that amino acids present in the extracellular domain just C-terminal of TMD1 are involved in ATP binding. Together, those findings suggested to us that the increased potencies of both ATP and αβmeATP seen for chimera 1.2.1 might originate from amino acids present in the rP2X$_2$-TMD1 region. To test this hypothesis, we constructed a new chimera that comprised the rP2X$_2$ subunit in which the only alteration was that its TMD1 domain was replaced by the TMD1 of rP2X$_2$ (this chimera is labeled TM1(r1)). As seen in Fig. 3a, this chimera also had increased sensitivity to both ATP and αβmeATP.

![Fig. 3. Replacement of only the first transmembrane domain of rP2X$_2$ confers changes in agonist, but not antagonist, sensitivities](image)

![Fig. 4. Agonist sensitivities of a chimeric receptor is determined by the first transmembrane domain.](image)
matched the reported values seen at the wt-rP2X2 and substan-
tially different from what has been observed for rP2X1 (20).

It was important to assess whether the data ob-
tained with the rP2X1-containing chimeras were due to rP2X1-
specific sequences, or whether results were indicative of P2X
receptor structure/function in general. We therefore con-
structed a chimera in which the TMD1 of rP2X2 was replaced
with that of the rP2X3 subunit (this chimera is labeled
TM1(r3)). rP2X3 was chosen because it is the only other rat
subunit at which αβmeATP has high affinity and potency.
As seen in Fig. 4a, both αβmeATP and ATP again displayed ap-
preciably increased potencies at TM1(r3) (ATP: EC50 = 0.39
μM; αβmeATP: EC50 = 8.9 μM) when compared with wt-P2X2.

To this point, results from the studies of agonists on the
various chimeras suggested that the agonist pharmacology of a
given receptor was determined, in large part, by the structure
of its TMD1. If this were true, then replacement of the P2X2
TMD1 with a TMD1 from a different αβmeATP-insensitive
subunit should not yield a chimeric protein with enhanced
agonist sensitivities. To test this hypothesis, we chose to use
the TMD1 from the zebrafish P2X3 (zP2X3) subunit as previous
work from our labs has demonstrated that αβmeATP has much
lower potency at zP2X3 (EC50 > 100 μM) than at rP2X2, while
having similar ATP sensitivity (EC50 = 1.5 μM) (21). Therefore,
a chimera, labeled TM1(z3), containing the TMD1 of the zP2X3
subunit transplanted into rP2X2 was constructed, and dose-
response curves for ATP and αβmeATP were determined. As
was predicted from our working model, this construct exhibited
ATP and αβmeATP sensitivities (Fig. 4b) (ATP: EC50 = 2.15
μM; αβmeATP: EC50 > 300 μM) similar to those of the wild-type
zP2X3 receptor.

Dependence of Agonist Efficacy on TMD1 Amino Acid Com-
position—Although the TM1(r3) and TM1(z3) chimeras had
quite different sensitivities to αβmeATP, their respective
TMD1 domains differed by only five residues (Fig. 5a). Thus,
a point mutation strategy was used in an effort to elucidate
which of the amino acid differences provided the structural
underpinnings of the pharmacological differences observed
between the two chimeras. In this approach, the chimera TM1(z3)
was used as the template in order that the end point (αβmeATP
sensitivity) would be a gain of function. We tested mutations
beginning with the N-terminal portion of the domain to deter-
mine which one(s) has a role in enhancing agonist potency. This
was done by comparing whole cell currents elicited by 30 and
100 μM αβmeATP to that of 30 μM ATP (a concentration that
elicits maximal whole cell currents). As seen in Fig. 5, the I34A
and L41S mutations did not show any increased sensitivity to
αβmeATP. Mutant C45G was not functional (data not shown),

The Effects of TMD1s from Other P2X Subunits on Agonist
Potencies—It was important to assess whether the data ob-

| subunit | TMD1 sequence | αβmeATP |
|---------|---------------|---------|
| rP2X2   | 34I34V,45G    | 0.3 nA  |
| zP2X3   | 34I34V,45G    | 0.3 nA  |

FIG. 5. No single amino acid difference between the TMD1 s of
the rat and zebrafish P2X2 subunits accounts for the differences
observed at the chimeras TM1(r3) and TM1(z3). a, a alignment of
the amino acid sequences present in the TMD1 regions of the rat (upper)
and zebrafish (lower) P2X2 subunits used to construct the chimeric
receptors; boxed residues are identical; αβmeATP sensitivity is indi-
cated by + or −. b, currents evoked from cells expressing the indicated
point mutations in the chimera TM1(z3). Traces shown are those in-
duced by 30 μM ATP (square), 30 μM αβmeATP (inverted triangle), and
100 μM αβmeATP (triangle), and are representative of those seen in
at least three cells for each mutation.

(EC50 = 1.3 μM) and αβmeATP (EC50 = 73 μM) that was
virtually indistinguishable from the values seen with 1.2.1, but
again much higher than those observed with wt-rP2X2. Thus,
TMD1 alone was sufficient to induce the increased potencies of
the two agonists observed at the 1.2.1 receptor.

To determine if the change observed in agonist potencies was
found for antagonists as well, we tested the actions of the
antagonist trinitrophenyl-ATP (TNP-ATP) on ATP-induced
currents in cells expressing TM1(r1). This compound has high
affinity and potency. As
shown in Fig. 3b, the affinity (IC50 = 1.7 μM) and the Hill slope (nH = −1.1) for
TNP-ATP inhibition of ATP-gated currents at TM1(r1) matched the reported values seen at the wt-rP2X2, and substan-
tially different from what has been observed for rP2X1 (20).
Thus, the effects of the TMD1 exchange were limited to the
action of agonists at the protein.

The Effects of TMD1s from Other P2X Subunits on Agonist
Potencies—It was important to assess whether the data ob-

FIG. 6. Triple mutation in TM1(z3) does not increase αβmeATP
potency to that seen at TM1(r3). Dose-response curves for ATP- and
αβmeATP-gated currents at TM1(z3) 144V/C45G/M49L performed as
described for TM1(r3). Values obtained from curve fits were ATP: EC50
= 1.5 ± 0.1 μM, nH = 1.3 ± 0.1; αβmeATP: EC50 = 155 ± 20 μM, nH =
1.5 ± 0.2.
resembling the single mutant M49L, and dose-response curves to that seen in TM1(r3). This triple mutant gave currents possible to increase the potencies of both membrane domain present in the monomer. By simply replacing meric receptors is derived from the structure of the first trans-

Values shown are the mean ± standard deviation; N = number of cells used for the determination; ND, could not be determined.

and therefore we made the double mutant I44V/C45G to compensate for the lethality of the single C45G mutation. This construct was functional but did not show an enhanced αβmeATP response either, thus effectively ruling out either residue as being essential for an enhanced sensitivity to agonists. Additionally, neither the double mutation I34A/L41S or residue as being essential for an enhanced sensitivity to ago-

Two possible mechanisms could explain these data. The first would be that the agonist binding site conformation had

| subunit | TM1 sequence | αβmeATP |
|---------|--------------|---------|
| rP2X1  | 29 HGVIFKLLTLLVNVVICVHVE 52 | +       |
| rP2X2  | 29 LIKIVFELLLVNVSCVHVEQ 52  | −       |
| rP2X3  | 23 M1TVFLLTVLVNVVCVIHE 46  | +       |
| rP2X4  | 28 VLLHFLLTVLVNVSCVIHE 51  | −       |
| rP2X5  | 29 VLLHFLLTVLVNVSCVIHE 51  | −       |
| rP2X6  | 30 VLSLVVLLVNVVCVIHE 53    | −       |
| rP2X7  | 26 VVLTLLHMTVFSFQFLA 49    | −       |

**Fig. 7. Alignment of the TM1s of all seven rat and zebrafish P2X subunit sequences. Boxed residues are indicate residues that exhibit identity across the TMD1 at the 65% level. αβmeATP sensitivity is indicated by a + or −.**

**Fig. 8. Effect of exchanging TMD2 in rP2X2 with that of rP2X1.** Dose-response curves for ATP- and αβmeATP-gated currents at TM2(r1). Values obtained from curve fits were ATP: EC_{50} = 0.72 ± 0.3 μM, n_H = 0.9 ± 0.3; αβmeATP: EC_{50} = 102 ± 36 μM, n_H = 0.72 ± 0.2.

changed as a result of the chimerism such that ATP and αβmeATP now interacted with higher affinity at the binding site present on the chimeras versus the wild type rP2X2. The second possibility would be that the transduction pathway linking agonist binding to opening of the channel gate had been altered in the chimeric protein such that a less precise fit of the agonist into the binding pocket could induce gating. As has been cogently argued (22), when performing mutational analyses of ligand-gated channels, it is extremely difficult to differentiate between mutational effects on agonist binding and those on channel opening, as both steps are dependent on, and affected by, one another. One approach to resolving this dilemma is to compare at least two agonists, differing in their efficacy, at each mutant receptor rather than comparing one mutant versus another using agonists of equal efficacy. This approach was used in our study, and, based on the findings, we do not favor the hypothesis that the observed increases in agonist potencies seen at a number of the chimeras resulted from changes only at the agonist binding site. Instead, the data provides a better fit to the hypothesis that the transduction properties of the receptor have been altered.

This interpretation is based on the following observations. First, ATP potency was shifted at the majority of the chimeras, whereas the αβmeATP response was increased only at a few constructs (summarized in Table I). Second, the αβmeATP response at 1.21 did not reach the maximum obtained with ATP, which suggests that it is acting as a partial agonist at this construct. However, at TM1(r1), TM1(r3), and the triple mutant TM1(z3) I44V/C45G/M49L, αβmeATP was as efficacious as ATP and thus was a full agonist at those constructs. Therefore, the findings from all the chimeras strongly imply that the effect of the chimerism on agonist potencies was mediated by an alteration in gating rather than a change in ligand binding. This argument is based on the logic that a mutation affecting

**TABLE I Summary of dose-response data**

| Construct | EC_{50} (μM) | n_H | N | EC_{50} (μM) | n_H | N |
|-----------|--------------|-----|---|--------------|-----|---|
| wt-rP2X2  | 22 ± 1       | 1.6 ± 0.1 | 8 | >300          | ND  | 6 |
| 1.2.1     | 0.93 ± 0.23  | 1.1 ± 0.3 | 7 | 49 ± 12       | 1 ± 0.3 | 8 |
| TM1(r1)   | 1.3 ± 0.2    | 1.7 ± 0.4 | 9 | 73 ± 18       | 1 ± 0.3 | 8 |
| TM1(r3)   | 0.39 ± 0.04  | 0.9 ± 0.07 | 5 | 8.9 ± 4.3     | 1.3 ± 0.7 | 7 |
| TM1(z3)   | 2.15 ± 0.3   | 1.6 ± 0.3 | 7 | <300          | ND  | 6 |
| TM1(z3)I44V/C45G/M49L | 1.5 ± 0.1 | 1.3 ± 0.1 | 10 | 155 ± 20 | 1.5 ± 0.2 | 7 |
| TM2(r1)   | 0.72 ± 0.3   | 0.9 ± 0.3 | 7 | 102 ± 36      | 0.72 ± 0.2 | 8 |

**DISCUSSION**

In this study, we provide evidence that the variation in ATP and αβmeATP potencies exhibited at various P2X homo-oligomeric receptors is derived from the structure of the first transmembrane domain present in the monomer. By simply replacing the TMD1 of an αβmeATP-insensitive subunit with the equivalent domains of a βmeATP-sensitive subunits, it was possible to increase the potencies of both αβmeATP and ATP in the chimeric receptor. Additional site-directed mutagenesis experiments indicated that this effect was not the direct result of any single amino acid substitution, but rather the product of a cumulative effect upon the overall structure of TMD1 induced by the multiple amino acid differences seen across individual subunit primary sequences.

This argument is based on the logic that a mutation affecting
binding only would be predicted to affect agonist potency but not alter efficacy at the mutant when compared with the parent receptor. On the other hand, if the gating efficiency (i.e. transduction) was affected, then different agonists could have differing efficacies at a mutant compared with the parent receptor due to intrinsic differences in their ability to induce channel gating; such a dependence of channel gating efficiencies on protein structure has been reported for other ligand-gated channels as well (23). A third finding was that the Hill slopes for ATP were shifted toward 1.0 for many of the chimeras (Table I), again a result that is easier to explain by a change in gating efficiency of a ligand rather than a simple change in binding affinity. Thus, the simplest interpretation of all of these results is that the outcome of transplanting the rP2X3 TMD1 into rP2X2 was an enhancement in the transduction, or gating, of the channel in response to occupancy of the agonist binding site rather than solely an increase in agonist affinity. This interpretation does not rule out the possibility that agonist affinity was also altered; it just excludes it as a sole reason for the observed results. In light of the lack of a high degree of conservation in the primary sequences of the P2X1 and P2X3 TMD1s, and across P2X subunits in general (Fig. 7), it would appear that the overall topological shape of TMD1, rather than a specific amino acid or two, is an important determinant of the agonist-induced gating behavior of the P2X receptor channel.

The findings presented here, when integrated with previous findings from our labs (8, 16) and others (12, 13), now provide a preliminary conceptual mechanism for understanding how ligand gating of the P2X receptor occurs, with TMD1 playing the role of a “trigger” in actuating channel gating. In this model, interaction of ATP with extracellular amino acid residues that are just C-terminal of TMD1 (and perhaps with other extracellular residues as well) would result in perturbations of structure that would have to be transmitted to the immediately adjacent proximal TMD1 region. This disturbance of TMD1 would then induce a local structural change that would influence TMD2, causing it to move in such a way as to allow opening of the gate and subsequent ion permeation. If this model is accurate, then it could be predicted that replacement of rP2X2 TMD2 with the TMD2 of rP2X1 should also yield a receptor (TM2(r1)) with altered agonist properties. As seen in Fig. 8, this is what was observed as both ATP and αβmeATP exhibited increased potencies (EC50 values of 0.72 and 102 μM, respectively) at this chimera. Interestingly, although αβmeATP is a full agonist at TM1(r1), it is only a partial agonist at TM2(r1), showing that the effect of an exchange of TMD2 on agonist-induced gating is not as robust as one of TMD1. It is tempting to speculate that this is because TMD2 has a less direct role in the transduction mechanism. Regardless, this result is in keeping with the hypothesis that TMD1 and TMD2 interact in the membrane, and that this interaction plays a role in the gating efficiencies of agonists at the protein.

Acknowledgments—We thank Laura Hobart and Olga Barmina for technical support.

REFERENCES

1. Friel, D. D., and Bean, B. P. (1988) J. Gen. Physiol. 91, 1–27.
2. Dubyak, G. R., and El-Moatassim, C. (1993) Am. J. Physiol. 265, C577–C606.
3. North, R. A., and Surprenant, A. (2000) Annu. Rev. Pharmacol. Toxicol. 40, 563–580.
4. Khakh, B. S., Burnstock, G., Kennedy, C., King, B. F., North, R. A., Seguela, P., Voigt, M., and Humphrey, P. P. (2001) Pharmacol. Rev. 53, 107–118.
5. Torres, G. E., Egan, T. M., and Voigt, M. M. (1999) J. Biol. Chem. 274, 6653–6659.
6. Evans, R. J., Lewis, C., Virgino, C., Lundstrom, K., Buell, G., Surprenant, A., and North, R. A. (1996) J. Physiol. 497, 413–422.
7. Rassendren, F., Buell, G., Newbolt, A., North, R. A., and Surprenant, A. (1997) EMBO J. 16, 3446–3454.
8. Egan, T. M., Haines, W. R., and Voigt, M. M. (1998) J. Neurosci. 18, 2350–2359.
9. Migita, K. M., Haines, W. R., Voigt, M. M., and Egan, T. M. (2001) J. Biol. Chem. 276, in press.
10. Buell, G., Lewis, C., Cillo, G., North, R. A., and Surprenant, A. (1996) EMBO J. 15, 55–62.
11. Garcia-Guzman, M., Soto, F., Gomez-Hernandez, J. M., Lund, P. E., and Stuhmer, W. (1997) Mol. Pharmacol. 51, 109–118.
12. Ennion, S., Hagan, S., and Evans, R. J. (2000) J. Biol. Chem. 275, 29361–29367.
13. Jiang, L. H., Rassendren, F., Surprenant, A., and North, R. A. (2000) J. Biol. Chem. 275, 34190–34196.
14. Burnstock, G. (1996) Obi Found. Symp. 198, 1–34.
15. Torres, G. E., Egan, T. M., and Voigt, M. M. (1998) FEBS Lett. 425, 19–23.
16. Haines, W. R., Voigt, M. M., Migita, K., Torres, G. E., and Egan, T. M. (2001) J. Neurosci. 21, in press.
17. Horn, R., and Korn, S. J. (1992) Methods Enzymol. 207, 149–155.
18. Haines, W. R., Torres, G. E., Voigt, M. M., and Egan, T. M. (1999) Mol. Pharmacol. 56, 720–727.
19. Le, R. T., Babinski, K., and Seguela, P. (1998) J. Neurosci. 18, 7152–7159.
20. Virginio, C., Robertson, G., Surprenant, A., and North, R. A. (1998) Mol. Pharmacol. 53, 969–973.
21. Egan, T. M., Cox, J. A., and Voigt, M. M. (2000) FEBS Lett. 475, 287–290.
22. Colquhoun, D. (1998) Br. J. Pharmacol. 125, 924–947.
23. O’Shea, S. M., and Harrison, N. L. (2000) J. Biol. Chem. 275, 22764–22768.
The First Transmembrane Domain of the P2X Receptor Subunit Participates in the Agonist-induced Gating of the Channel
William R. Haines, Keisuke Migita, Jane A. Cox, Terrance M. Egan and Mark M. Voigt

J. Biol. Chem. 2001, 276:32793-32798.
doi: 10.1074/jbc.M104216200 originally published online July 3, 2001

Access the most updated version of this article at doi: 10.1074/jbc.M104216200

Alerts:
- When this article is cited
- When a correction for this article is posted

Click here to choose from all of JBC's e-mail alerts

This article cites 21 references, 12 of which can be accessed free at http://www.jbc.org/content/276/35/32793.full.html#ref-list-1