Thyroid hormone regulates postnatal expression of transient K^+ channel isoforms in rat ventricle

Y. Shimoni, C. Fiset, R. B. Clark, J. E. Dixon*, D. McKinnon* and W. R. Giles†

*Departments of Physiology and Biophysics and Medicine, University of Calgary School of Medicine, Calgary, Alberta, Canada T2N 4N1 and *Department of Neurobiology and Behavior, State University of New York, Stony Brook, NY 11794-5230, USA*

1. The ability of thyroid hormone to regulate the postnatal changes of the Ca^{2+}-independent transient outward K^+ current (I_t) was studied in rat ventricular myocytes.

2. In rat ventricle, I_t is very small at birth and then increases markedly between postnatal days 8 and 20. The time course of this increase in current density is similar to that of a significant rise in plasma thyroid hormone (T_3) levels.

3. During early development, the density of expression of I_t can be altered by changes in thyroid hormone levels. Eight days after birth the density of I_t measured at +50 mV in control animals is $2{\cdot}2 \pm 0{\cdot}4$ pA pF^{-1}. This value is about 3-fold larger ($6{\cdot}5 \pm 0{\cdot}8$ pA pF^{-1}) in myocytes from age-matched hyperthyroid animals. When the plasma T_3 level in newborn rats is not allowed to increase, or is decreased by making animals hypothyroid, this age-dependent increase in I_t fails to occur.

4. Using RNase protection assays, $K_v4.2$ and $K_v4.3$ mRNA levels were measured in ventricular tissues obtained from age-matched 8-day-old control and hyperthyroid rats. In hyperthyroid animals, where an approximately 3-fold increase in I_t was identified, increases in the mRNA levels for $K_v4.2$ and $K_v4.3$ were 1·6-fold and 2·6-fold, respectively.

5. These results show that thyroid hormone can regulate the development of I_t in rat ventricle. Direct measurements of I_t density and mRNA levels as a function of development and thyroid hormone levels also strongly suggest that the $K_v4.2$ and $K_v4.3$ channels are essential components of I_t in rat ventricular cells.

In many mammalian species, including man, the Ca^{2+}-independent transient outward K^+ current, I_t (Campbell, Rasmusson, Comer & Strauss, 1995; Giles, Clark & Braun, 1996) exhibits postnatal increases in both ventricular and atrial tissue (Kilborn & Fedida, 1990; Jeck & Boyden, 1992; Sanchez-Chapula, Elizalde, Navarro-Polanco & Barajas, 1994; Wahler, Dollinger, Smith & Flemal, 1994; Crumb, Pigott & Clarkson, 1995). In adult rabbit ventricle the expression level (density) of this current has also been shown to be very sensitive to elevated thyroid hormone (T_3) levels (Shimoni, Banno & Clark, 1992). In contrast, in adult rat ventricle elevated T_3 levels do not change the size of I_t (Shimoni, Severson & Giles, 1995).

The rat is born with a poorly developed hypothalamic–pituitary–thyroid axis, and the final stage of its maturation occurs postnatally (Vigouroux, 1976). Earlier work (Fisher, Dussault, Sack & Chopra, 1977) showed that a significant increase in T_3 occurs between postnatal days 10 and 20. This is also the time period during which I_t exhibits the most rapid increase (Wahler *et al.* 1994). We therefore tested the hypothesis that there is a causal relationship between the postnatal surge in T_3 levels and the increase of I_t. In these experiments, the postnatal expression of I_t and changes in T_3 levels were directly compared; in addition, the thyroid status of newborn rat pups was manipulated to determine whether this alters the size of I_t during development. In rat heart, this type of intervention is known to affect the postnatal development of noradrenergic neurotransmission (Lau & Slotkin, 1982; Metz, Seidler, McCook & Slotkin, 1996), myosin isozyme composition (Chizzonite & Zak, 1984), the expression levels of selected isoforms of the Na^+–K^+-ATPase (Orlowski & Lingrel, 1990; Ewart & Klip, 1995), as well as L-type Ca^{2+} channels and Ca^{2+}-ATPase (Wibo, Kolar, Zheng & Godfraind, 1995).

A number of different K^+ channels cloned from heart tissue can generate Ca^{2+}-independent transient outward K^+ currents in heterologous expression systems. These include $K_v1.4$, $K_v4.2$ and, as has been shown very recently, $K_v4.3$

† To whom correspondence should be addressed.

Table 1. Mean [T_3]

Postnatal days	[T_3] (nM) Euthyroid group	Hyperthyroid group
6	0·65	—
8	0·87	2·0
12	1·15	3·0
16	1·50	2·8
20	1·52	5·8
35	1·50	—

(Deal, England & Tamkun, 1996; Barry & Nerbonne, 1996; Dixon *et al.* 1996). It has been suggested previously, based on mRNA expression studies, that both the $K_v4.2$ and $K_v4.3$ channels underlie the I_t in rat ventricle (Dixon & McKinnon, 1994; Dixon *et al.* 1996). Our observation that antisense oligonucleotides directed against either the $K_v4.2$ or $K_v4.3$ mRNA can reduce expression of I_t in rat ventricular myocytes is consistent with this hypothesis (Fiset, Clark, Shimoni & Giles, 1997). In further support of the possibility that these two K_v4 channels are the primary contributors to the I_t is the observation made by Roberds & Tamkun (1991) that $K_v1.4$ mRNA expression is low in adult rat ventricle. It has also been reported that $K_v1.4$ protein is expressed at very low levels in ventricular myocytes (Barry, Trimmer, Merlie & Nerbonne, 1995) whereas the $K_v4.2$ channel is relatively abundant. Pharmacological evidence also favours a functional role of $K_v4.2$ in rat ventricle. The transient outward K^+ current I_t is blocked by low concentrations of the antiarrhythmic agent, flecainide. In contrast, the current due to $K_v1.4$ channel expression is not altered by flecainide (Dixon *et al.* 1996; Yeola & Snyders, 1997).

In the present experiments we have compared the density of I_t with the mRNA levels for $K_v1.4$, $K_v4.2$, and $K_v4.3$ in ventricles from 8-day-old rat pups in control and hyperthyroid conditions. Our results show that I_t expression and mRNA levels for $K_v4.2$ and $K_v4.3$ are strongly modulated by T_3 levels in neonatal rat ventricle, and support the hypothesis that the $K_v4.2$ and $K_v4.3$ channels have a significant functional role in the expression of this Ca^{2+}-independent transient outward current in this tissue.

METHODS

Single cell preparation

Myocytes from the ventricles of adult and neonatal rats were obtained by enzymatic dispersion as described in detail in the companion (preceding) paper (Fiset *et al.* 1997).

Electrophysiological and data recording procedures

The methods for microelectrode preparation, voltage clamp recordings and data analyses were identical to those described in detail in Fiset *et al.* (1997).

Thyroid status

Hyperthyroid conditions were produced in newborn pups by daily subcutaneous injections of triiodothyronine (T_3, 0·2 μg (g body weight)$^{-1}$). Hypothyroid conditions were established by daily injections of an antithyroid drug, propylthiouracil (PTU, 20 μg (g body weight)$^{-1}$ given subcutaneously). In addition, pregnant females received PTU (0·05%) in their drinking water 4–5 days prior to delivery, and throughout the postnatal period. This rendered these females hypothyroid (as confirmed by measuring plasma T_3 levels) and minimized T_3 transfer from them through the placenta or in the milk in the pre- and postnatal periods, respectively. Thyroid status was confirmed by collecting plasma from control, hyper- and hypothyroid pups, and measuring T_3 levels using standard radioimmunoassay methods (Foothills Hospital Clinical Laboratory, Calgary, Canada). The mean T_3 values for our experimental groups are shown in Table 1.

These control (euthyroid) values and the postnatal changes in them are very similar to those reported by Chizzonite & Zak (1984) and Fisher *et al.* (1977). All the animals in the hypothyroid group had T_3 levels of less than 0·40 nM, which was the lower limit of detection of the radioimmunoassay which was used.

RNase protection assay

A RNase protection assay was used to compare the mRNA levels for $K_v1.4$, $K_v4.2$ and $K_v4.3$ K^+ channel isoforms in 8-day-old rat ventricles obtained from control and hyperthyroid animals. The preparation of the RNA and the RNase protection assay were performed as described previously by Dixon & McKinnon (1994). Briefly, total RNA was prepared from rat ventricles obtained from 8-day-old rats (control and hyperthyroid). Hearts were rapidly excised and the ventricles washed in normal Tyrode solution. Ventricles were then quickly frozen in liquid nitrogen. For each RNA sample, tissue from five hearts was pooled and homogenized in guanidinium thiocyanate. Total RNA was prepared by pelleting the homogenate over a CsCl step gradient. RNA samples were quantified by spectrophotometry, and then rediluted to give approximately equal concentrations of RNA in each sample. These normalized samples were then re-measured by spectrophotometry to confirm that all samples had similar concentrations of mRNA, and this stock was used for subsequent experiments. This procedure served to minimize errors in the amount of RNA in each test sample due to inaccurate pipetting volumes. Ten micrograms of total RNA were used for the RNase protection assay. The sequence of the cDNA templates used for the production of the radioactive RNA probes for $K_v1.4$, $K_v4.2$ and $K_v4.3$ have been reported previously (Dixon & McKinnon, 1994; Dixon *et al.* 1996). Specific RNA expression was quantified using a PhosphorImager (Molecular Dynamics Inc., Sunnyvale, CA, USA).

Statistics

Data are reported as means ± S.E.M. Student's two-tailed, unpaired t test was used to evaluate any significant differences between different groups. A t value giving $P < 0·05$ was considered significant.

RESULTS

Time course of development of I_t in neonatal rat ventricle

Figure 1 shows the time course of postnatal development of the Ca^{2+}-independent transient outward current (I_t) in rat ventricle. In Fig. 1*A* the four sets of superimposed current records demonstrate the changes in I_t at these selected

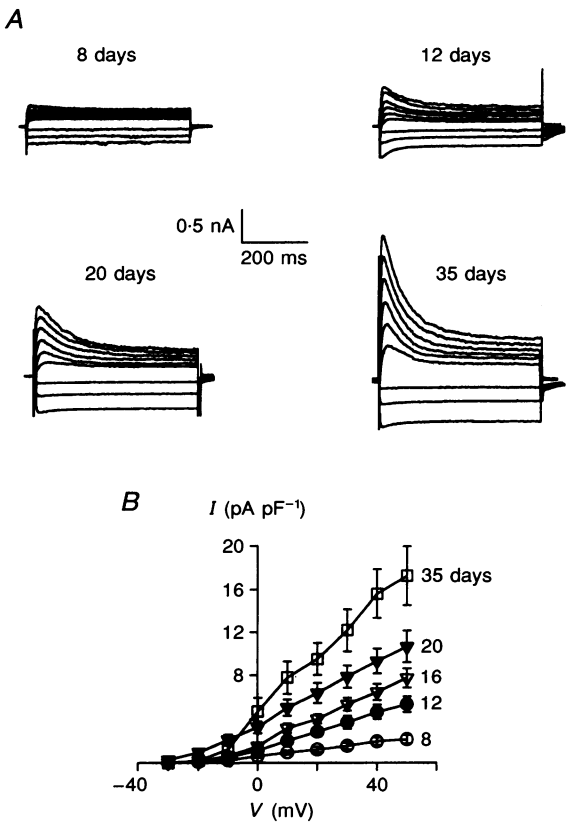

Figure 1. Postnatal development of the Ca^{2+}-independent transient outward K^+ current, I_t, in rat ventricular myocytes

A, each of the four sets of superimposed current traces were recorded from ventricular myocytes in response to 500 ms hyperpolarizing and depolarizing steps applied at 0·1 Hz from the holding potential (−80 mV) to −110, −100 and −90 mV; and in 10 mV steps from 0 mV to 50 mV. Cells obtained from neonates at 8, 12, 20 and 35 days of age were studied. *B*, corresponding current densities from groups of cells ($n = 20$–45) at the same postnatal stages as in *A*. Temperature in these and all other recordings was 23 ± 1 °C.

stages of development. The current–voltage relationships in Fig. 1*B* were obtained from ventricular myocytes at postnatal days 8, 12, 16, 20 and 35. These results demonstrate the rapid development of I_t early in the postnatal period as has been described previously (Kilborn & Fedida, 1990; Wahler *et al.* 1994), and also show that the density of the background inwardly rectifying K^+ current (I_{K1}) does not change significantly between days 8 and 35. Note that although the inward current due to I_{K1} is larger in the day 35 myocyte than in the day 8 cell, this increase can be completely accounted for by the increase in capacitance as the cell increases in size during development. In these experiments I_{K1} was activated by 500 ms hyperpolarizing voltage clamp steps to −90, −100 and −110 mV. The data in Fig. 2 show that the background inward rectifier K^+ current, I_{K1} (measured in picoamps per picofarad), does not change significantly between postnatal days 8 and 20. These results also demonstrate that the rapidly activated slowly inactivating K^+ current in this tissue (I_{sus}) remains unchanged.

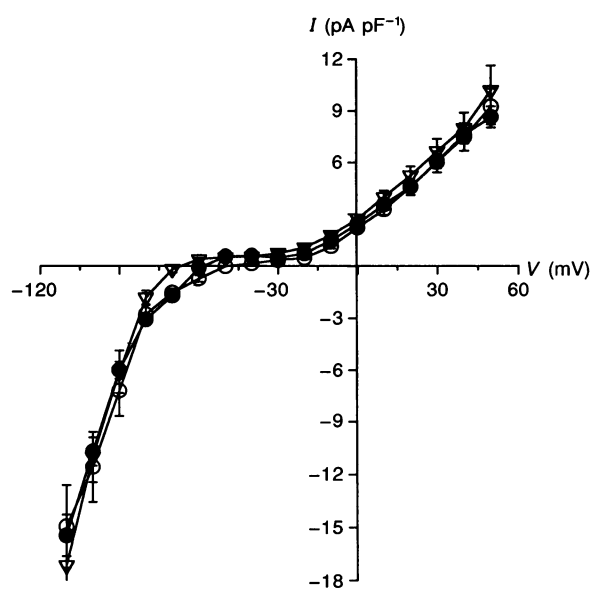

Figure 2. Postnatal changes in the background inwardly rectifying K^+ current (I_{K1}) and in the rapidly activating and slowly inactivating outward K^+ current (I_{sus}) in rat ventricle.

Mean current–voltage relationships for I_{K1} and I_{sus} activated by voltage-clamp steps ranging from −110 mV to +50 mV in 10 mV increments from the holding potential −80 mV. Cells were studied from neonates at 8 (○), 16 (●) and 20 days of age (▽).

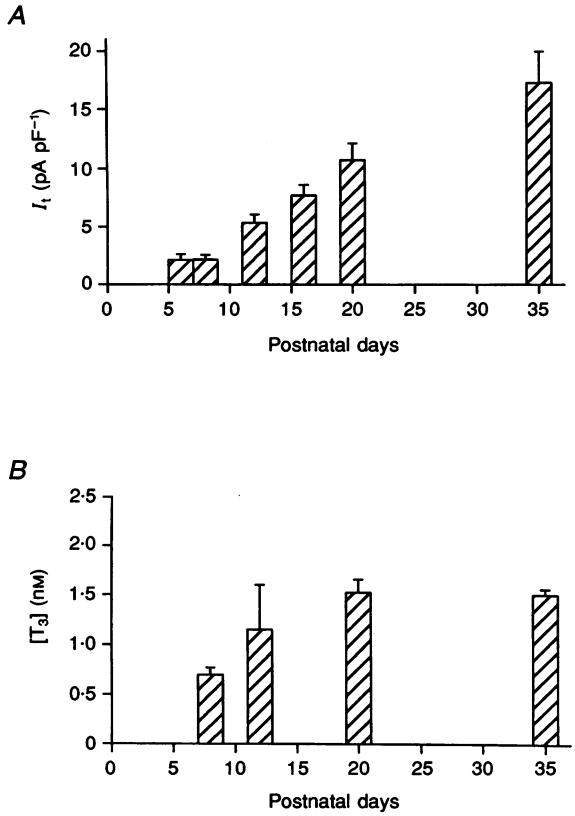

Figure 3. Postnatal changes in I_t and thyroid hormone levels in rat ventricle

Age-dependent development of I_t measured at +50 mV is shown in *A*, and changes in plasma thyroid hormone (T_3) levels are shown in *B*.

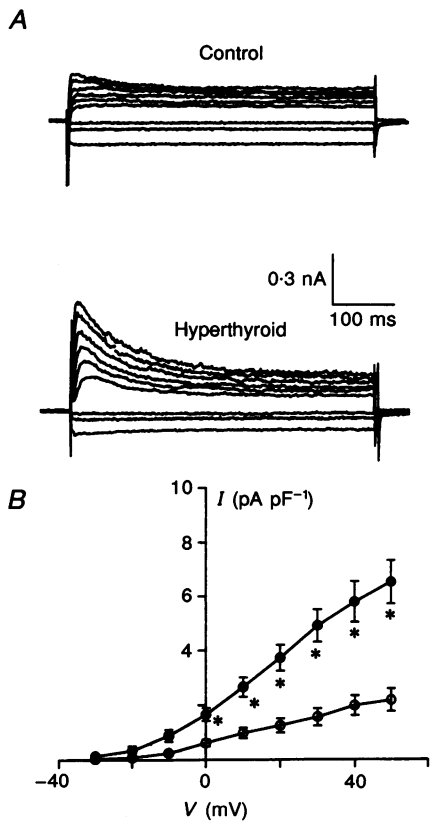

Figure 4. Effects of hyperthyroid conditions on I_t in neonatal rat ventricle

A, superimposed current traces recorded from ventricular myocytes of 8-day-old neonates with the same voltage-clamp protocol used in Fig. 1. The currents at the top are from a control animal, and those below are from a rat pup made hyperthyroid (see Methods). *B*, current–voltage relationships showing mean current densities obtained from two groups (control, ○, $n = 28$; hyperthyroid, ●, $n = 36$) of 8-day-old neonates. * $P < 0{\cdot}05$. I_t density is increased significantly ($P < 0{\cdot}001$) by the hormone treatment.

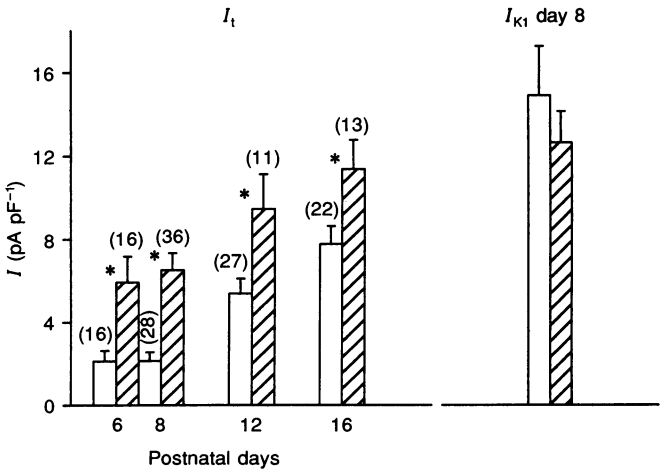

Figure 5. Acceleration of I_t development in neonatal ventricular myocytes by T_3 hormone treatment

Mean values of I_t at different ages are shown for untreated (□) and hormone-treated (▨) animals. * $P < 0{\cdot}05$. T_3 treatment very significantly enhanced I_t in early development (6- and 8-day-old pups), and had relatively smaller effects at days 12 and 16. T_3 treatment had no significant effect on I_{K1} in 8-day-old pups. The number of cells studied at each development stage is given in parentheses.

Figure 3 compares the time course of postnatal increase in I_t with changes in plasma thyroid hormone (T_3) levels as a function of neonatal development. These pooled results show that substantial increases in both I_t and T_3 occur between days 8 and 20. Thereafter, T_3 levels remain almost unchanged while I_t continues to increase.

Time course of development of I_t following altered thyroid status

The regulation of I_t by T_3 levels during neonatal development was studied in more detail by making newborn rats hyperthyroid with daily injections of T_3 (see Methods). Direct comparisons between the density of I_t in age-matched control and hyperthyroid animals were based upon data obtained at 6, 8, 12 and 16 days. In each set of measurements half of a litter of newborn rat pups were injected once per day with T_3 while the other half served as the controls. At selected time points (e.g. 8 or 16 days) ventricular myocytes were isolated from both control and treated animals, and electrophysiological experiments were carried out in the next 4–6 h.

The hyperthyroid state (a 2- to 3-fold increase in plasma T_3 levels) caused a marked increase in the size of I_t at all developmental stages which were studied. Figure 4 shows representative I_t records from control and T_3-treated 8-day-old animals. The pooled data in Fig. 4, plotted as current–voltage relationships, show that I_t in cells from hyperthyroid animals was significantly larger ($P < 0{\cdot}05$)

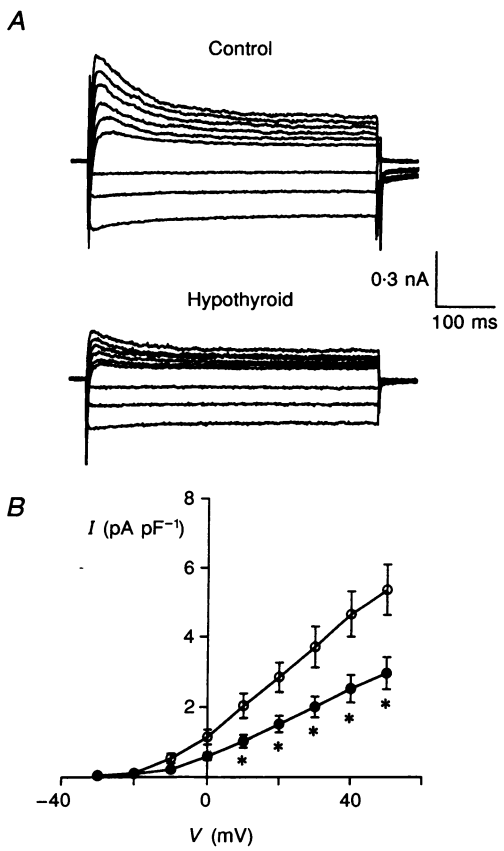

Figure 6. Effects of hypothyroid conditions on I_t in myocytes from neonatal rat ventricles

A, superimposed current traces recorded from ventricular myocytes of 12-day-old neonates with the same voltage-clamp protocol used in Fig. 1. Control denotes currents from an untreated animal; the bottom traces are from a rat pup made hypothyroid (see Methods). *B*, mean current densities for these two experimental groups (control, ○, $n = 27$; hypothyroid, ●, $n = 48$) show a very significant attenuation of I_t in the hypothyroid group. * $P < 0{\cdot}05$.

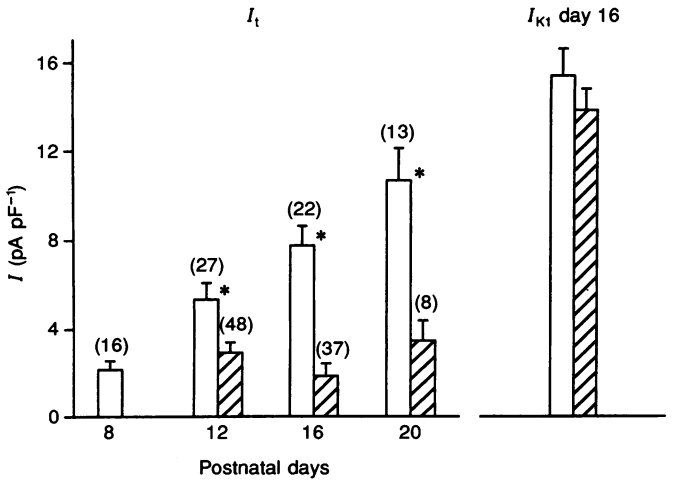

Figure 7. Expression of I_t is reduced under hypothyroid conditions in neonatal rat ventricle
□, mean I_t density at +50 mV for untreated (control) myocytes at days 8–20. ▨, mean I_t values for myocytes obtained from hypothyroid rat pups. $^*P < 0.05$. Hypothyroidism significantly decreases I_t at these developmental stages (days 12–20). I_{K1} was not significantly effected by hypothyroid conditions in 16-day-old pups. The number of cells studied at each developmental stage is given in parentheses. Measurements of I_{K1} were made from 22 control cells, and 34 cells from hypothyroid animals.

than that in age-matched control myocytes over a broad range of membrane voltages. A very similar pattern of results was obtained when this same comparison was done at the 6, 8, 12 and 16 day developmental stages (Fig. 5). In contrast, previous results have shown that similar T_3 levels in adult rats did not result in a significant increase in I_t (Shimoni *et al.* 1995). Thus, in rat ventricle hyperthyroid conditions result in significant increases in I_t early in neonatal development (e.g. between days 6 and 16), but this augmentation does not occur in adult ventricle. The right-hand panel of Fig. 5 shows I_{K1} data from 8-day-old rat pups. These results demonstrate the lack of effect of T_3 on I_{K1}.

An opposite pattern of results was obtained in ventricular cells from neonatal rat pups which had been made hypothyroid (see Methods). As shown in Fig. 6, I_t density at day 12 was significantly smaller in myocytes from hypothyroid animals than in the age-matched controls. The pooled results in Fig. 7 compare I_t densities at +50 mV in control and hypothyroid animals at days 12, 16 and 20. These results demonstrate that reduction of T_3 levels decreases I_t at each of these developmental stages, without any significant changes in I_{K1} (Fig. 7, right).

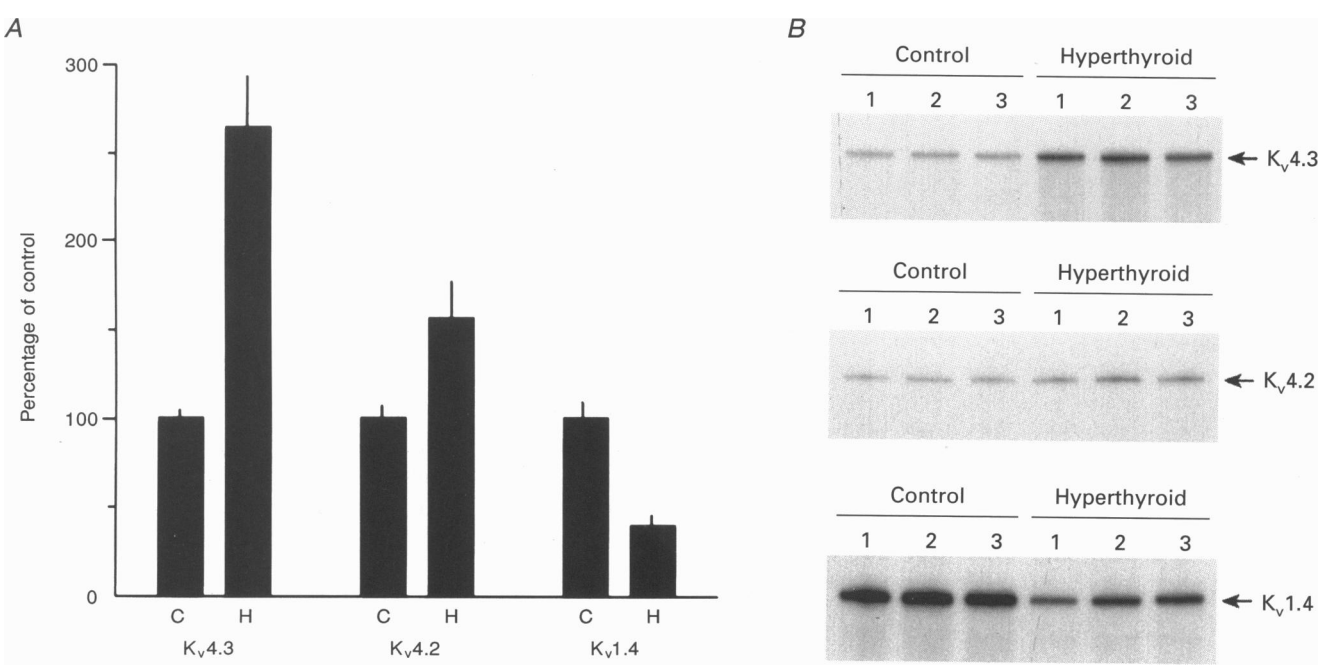

Figure 8. Comparison of $K_v1.4$, $K_v4.2$ and $K_v4.3$ mRNA levels in rat ventricles obtained from 8-day-old control and hyperthyroid animals
A, relative abundance of these three transcripts in control (C) and hyperthyroid (H) rats, determined by RNase protection assay. Means ± S.D. are shown. *B*, mRNA levels for the three K^+ channel isoforms measured in 3 control and 3 hyperthyroid samples (5 hearts per sample).

mRNA levels for K^+ channel isoforms in rat ventricle

RNase protection assays were used to assess $K_v1.4$, $K_v4.2$ and $K_v4.3$ mRNA levels in tissue from the ventricles of control and T_3-treated 8-day-old rat pups. The results from this set of experiments demonstrated that T_3 treatment significantly increased the mRNA levels for $K_v4.2$ and $K_v4.3$ and decreased the level of $K_v1.4$ mRNA expression in hyperthyroid animals (Fig. 8). The increase in $K_v4.3$ mRNA relative to controls was $2{\cdot}6 \pm 0{\cdot}3$-fold ($P < 0{\cdot}001$) and the increase in $K_v4.2$ mRNA was $1{\cdot}6 \pm 0{\cdot}2$-fold ($P < 0{\cdot}02$). The decrease in $K_v1.4$ mRNA was to a level of $0{\cdot}4 \pm 0{\cdot}07$ ($P < 0{\cdot}001$) compared with controls. In the 8-day-old control animals the $K_v4.3$ mRNA was approximately 1·7-fold more abundant that the $K_v4.2$ mRNA suggesting that the $K_v4.3$ channel may be expressed at higher levels at this stage of development. These results support the hypothesis that $K_v4.2$ and $K_v4.3$ channels underlie I_t in rat heart and demonstrate that the $K_v1.4$ channel makes little or no contribution to this current.

DISCUSSION

Summary of new findings

The results from these experiments demonstrate that physiologically relevant levels of thyroid hormone, T_3, can significantly alter the density of expression of a Ca^{2+}-independent transient outward K^+ current in neonatal rat ventricle. Our hypothesis that T_3 levels could modulate I_t density was based mainly upon two previously reported sets of data in which the time course of changes in I_t during postnatal development (Wahler *et al.* 1994) and the postnatal surge in T_3 levels (Vigouroux, 1976; Fisher *et al.* 1977) were described. One previous electrophysiological study in adult guinea-pig ventricle (Felzen, Rubinstein, Lotan & Binah, 1991) drew attention to the relationship between thyroid hormone levels and alterations in action potential shape. The relationship between plasma T_3 levels and density of I_t expression was demonstrated convincingly in our experiments by alterations in T_3 levels at selected stages of development. Thus, hyperthyroid conditions (Figs 4 and 5) augment the expression of I_t, and hypothyroid conditions diminish the expression of I_t (Figs 6 and 7). These T_3 effects on I_t appear to be quite selective: two different K^+ currents, the background inward rectifier (I_{K1}) and a sustained K^+ current (I_{sus}) were not changed when T_3 levels were altered.

Our results also provide an indication that the effects of T_3 on I_t are most prominent, or may occur exclusively, during postnatal development, since elevation of T_3 level had no significant effect on I_t in adult rat ventricle (Shimoni *et al.* 1995; Fig. 5). A somewhat similar, developmentally regulated, effect of T_3 on the expression of the α-subunit of the Na^+–K^+-ATPase in rat ventricle has been reported by Schmitt & McDonough (1988); Kamitani *et al.* (1992); and Zahler, Sun, Ardito & Kashgarian (1996). In addition, Metz *et al.* (1996) have described an effect of T_3 on the ontogenesis of the α-adrenergic receptor system in rat heart. All of these T_3-mediated effects also occur mainly early in postnatal development.

Previous work in a number of different laboratories has attempted to identify the K^+ channel isoforms that underlie the transient outward K^+ current in rat ventricle (Dixon & McKinnon, 1994; Barry *et al.* 1995; for review see Deal *et al.* 1996; Barry & Nerbonne, 1996). Our work (Dixon & McKinnon, 1994; Dixon *et al.* 1996; Fiset *et al.* 1997), and that of others (Blair, Roberds, Tamkun & Hartshorne, 1991; Serôdio, Vega-Saenz de Meira & Rudy, 1996), has provided evidence that the K^+ channel isoforms $K_v4.2$ and $K_v4.3$ are essential components of the K^+ channels that underlie I_t in rat and canine ventricles. The present electrophysiological studies and complementary molecular biology experiments involving the modulation of I_t by selected T_3 levels provide an additional means of evaluating this hypothesis. These results (Fig. 8) provide strong evidence that both the $K_v4.2$ and $K_v4.3$ genes play an important role in the control of expression of I_t in neonatal rat ventricle.

Relation to previous work

Thyroid hormone can regulate cardiac function by altering heart rate, velocity of contraction, and rate of diastolic relaxation (Morkin, Flink & Goldman, 1983). Moreover, each or a combination of these parameters can change during the pathophysiological states associated with either hyper- or hypothyroidism (Franklyn & Gammage, 1996). Work within the past 10 years has provided some significant insights into the cellular changes which underlie some of these alterations (Izumo, Nadal-Ginard & Mahdavi, 1986; Brent, Moore & Larsen, 1991; Ewart & Klip, 1995). It is now well known that thyroid hormone can regulate the time course and the extent of expression of specific myosin isozymes (Chizzone & Zak, 1984), thus providing part of the explanation for the altered rate of contraction. In addition, a number of the steps in excitation–contraction coupling and Ca^{2+} homeostasis in the rat myocardium are known to be modulated by plasma T_3 levels. These include the Ca^{2+}-ATPase, the density of expression of L-type Ca^{2+} channels as measured by ligand binding, and the ontogenesis of the sarcoplasmic reticulum (Wibo, Kolar, Zheng & Godfraind, 1995). In rat heart the expression of the Na^+–K^+-ATPase and Na^+–Ca^{2+} exchanger are modulated in a reciprocal fashion by T_3 levels (Magyar, Wang, Azuma & McDonough, 1995). The concentration–effect relationship for these changes has an EC_{50} of approximately 1 nM (Orlowski & Lingrel, 1990; Kamitani *et al.* 1992), which is similar to the apparent dose sensitivity which we have observed. Additional work will be needed to determine how this wide-ranging programme of changes in cellular microanatomy, enzymatic content and activity, and ion channel expression act in concert to bring about the physiological and pathophysiological alterations due to T_3 levels in the mammalian myocardium (Ewart & Klip, 1995).

Previous work has demonstrated hormone-dependent changes in the expression of a different cardiac K^+ channel. Takimoto & Levitan (1994) reported that glucocorticoid can upregulate $K_v1.5$ channel gene expression in rat ventricle.

Limitations of experimental design and interpretation of data

Thyroid hormone metabolism and its mechanism(s) of action have been studied very extensively in both neonatal and adult rat heart models. However, in some respects T_3 effects in rat heart are quite different from those in other mammals (Morkin *et al.* 1983). It will therefore be necessary to demonstrate the modulation of I_t by T_3 in the hearts of other mammalian species before concluding that the T_3 effects which we have observed are widely applicable. Our results establish a correlation between changes in plasma T_3 levels and the density of expression of I_t; and they also demonstrate that alterations in T_3 levels are associated with significant changes in mRNA levels for selected K^+ channel isoforms. Since thyroid hormone has many different effects on neonatal and adult heart (Metz *et al.* 1996), it is possible that the effects of T_3 on mRNA levels for $K_v4.2$ and $K_v4.3$ are indirect. For example, T_3 alters the number of adrenergic receptors in these preparations (Lau & Slotkin, 1982; Metz *et al.* 1996). More convincing evidence demonstrating a direct cause-and-effect relationship between T_3 and the expression of $K_v4.2$ and $K_v4.3$ could perhaps be obtained by using direct bath application of defined T_3 levels on cultured myocytes from neonatal rat ventricle (Kansara, Mehra, von Hagen, Kabotyansky & Smith, 1996). However, it has been reported recently that K_v α-subunit protein expression levels do not always correlate with mRNA levels in rat heart (Xu *et al.* 1996).

Our data demonstrate a striking modulation of I_t in neonatal rat heart by T_3 hormone levels which were altered using experimental protocols known to establish hypo- and hyperthyroid conditions in rat models (Oppenheimer, Silva, Schwartz & Sarks, 1977). These electrophysiological changes occur within the developmental time periods and the concentration ranges previously established for a number of other prominent T_3-induced enzyme changes in rat heart (Kolar *et al.* 1992; Orlowski & Lingrel, 1992). In summary, the results from our electrophysiological measurements and RNase assays demonstrate that alterations in T_3 levels, presumably triggered by changes in T_3-induced transcription or translational events, result in selective alteration of potassium channel isoforms thought to underlie I_t, namely $K_v4.2$ and $K_v4.3$.

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