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FACTORS INFLUENCING PLASMA PHENOBARBITONE LEVELS IN EPILEPTIC PATIENTS

M.J. EADIE, C.M. LANDER, W.D. HOOPER & J.H. TYRER
Department of Medicine, University of Queensland, Royal Brisbane Hospital, Brisbane 4029, Australia

1 Various statistical techniques were used to study the effects of age, sex and concurrent therapy with other anticonvulsants on the relation between plasma phenobarbitone levels and doses of (i) phenobarbitone, (ii) methylphenobarbitone or (iii) primidone, in epileptic patients.

2 Methylphenobarbitone and primidone are converted to phenobarbitone in the body. The mean doses of phenobarbitone, methylphenobarbitone and primidone which produced the same plasma phenobarbitone level (15 μg/ml) were, respectively, 1.75, 2.75 and 7.75 mg kg^{-1} day^{-1}.

3 For both phenobarbitone and methylphenobarbitone dose requirement to achieve a given plasma phenobarbitone level fell progressively with age. Sex influenced the relation between plasma phenobarbitone level and phenobarbitone or methylphenobarbitone dose. Interactions were detected between primidone and both phenytoin and carbamazepine.

4 In individual patients, within the limits of dosage studied, the relation between plasma phenobarbitone level and drug dose was not rectilinear if phenobarbitone itself was taken, but was rectilinear if methylphenobarbitone was taken.

Introduction

The anticonvulsant action of phenobarbitone has been known since 1912 (Hauptmann, 1912). Usually phenobarbitone itself is prescribed for patients, but the drug may also be provided indirectly by prescribing methylphenobarbitone or primidone. In animals each of these latter substances appears to be an anticonvulsant in its own right (Craig & Shideman, 1971; Gallagher, Smith & Mattson, 1970). However, each is in part biotransformed to phenobarbitone, and this is more slowly eliminated than the parent drugs. Consequently in the steady state after regular dosage, plasma phenobarbitone levels come to be higher than plasma levels of methylphenobarbitone or primidone (Butler & Waddell, 1958; Booker, Hosokowa, Burdette & Darcey, 1970). The derived phenobarbitone may be responsible for much of the anticonvulsant action of methylphenobarbitone or primidone. There has been some study of the effect of various factors on steady state phenobarbitone levels in patients taking the phenobarbitone itself (Buchthal & Lennox-Buchthal, 1972; Travers, Reynolds & Gallagher, 1972), and in patients taking primidone (Travers *et al.*, 1972). However, very little has been published on plasma phenobarbitone levels in patients taking methylphenobarbitone. Therefore the effects of various factors which might influence plasma phenobarbitone levels were studied in patients taking phenobarbitone, methylphenobarbitone, or primidone.

Methods

Since 1970 plasma phenobarbitone levels have been measured as a service for clinicians managing patients with epilepsy at the Royal Brisbane Hospital. At first a spectrophotometric technique was used (Wallace, 1969), but more recently levels have been measured by gas-liquid chromatography after forming a butyl derivative (Hooper, Dubetz, Eadie & Tyrer, 1975). The great majority of estimations were made by the latter technique, but results were very similar when the two assays were applied to the same plasma samples. From the accumulated data, records were extracted for all patients taking phenobarbitone, methylphenobarbitone or primidone and in whom there was adequate information as to age, sex, body weight, and dosage of phenobarbitone, or methylphenobarbitone or primidone and of any concurrent anticonvulsant therapy. The information was not analysed unless the patient had taken a constant dose of phenobarbitone (or congener) for at least 4 weeks, so that patients should have been in the steady state as regards drug intake and output (the elimination half-life of phenobarbitone is about 4 days (Mark, 1963).

Data were analysed for 121 patients (58 males, 63 females) taking phenobarbitone (made by various manufacturers), for 123 patients (61 males, 62 females) taking methylphenobarbitone (Winthrop Laboratories), and for 58 patients (35 males, 23 females) taking primidone (Imperial Chemical

Industries). The drugs were taken two or three times a day, except for a few adults who took a single daily dose of methylphenobarbitone in the evening. For a given patient only a single measurement of plasma phenobarbitone concentration at any one dosage level was studied. Where a patient had plasma phenobarbitone levels measured while taking phenobarbitone (or a congener), at different dosages, a single measurement at every dosage level was included in the study.

Using a Hewlett Packard programmable desk calculator, rectilinear and various types of curvilinear regression (exponential, power, second degree polynomial) were fitted to the data for the relation between plasma phenobarbitone level and doses of the three drugs studied (expressed on a body weight basis). The effects of age and sex on the linear regressions for plasma phenobarbitone level on drug doses were examined by covariance analysis techniques. In studying the effects of age, the population was divided on an age basis into four groups of reasonably equal numbers. If the relation between plasma drug level and dose differed between two neighbouring groups, each group was subdivided. If the relation did not differ, the two groups were fused into one. This process continued till it was no longer possible to define any further age groups which differed to a statistically significant degree from their neighbouring age groups. Multi-variable linear regression analysis was used to evaluate the effects of dosage of phenytoin, carbamazepine and sulthiame (drugs commonly prescribed in conjunction with the drugs under study) on the relation between plasma phenobarbitone levels and dose of phenobarbitone or methylphenobarbitone or primidone. The numbers of patients taking these non-barbiturate anticonvulsants are shown in Table 1. The relation between plasma phenobarbitone level and drug dose was studied by graphical techniques in individual patients who took phenobarbitone, methylphenobarbitone or primidone in different doses at different times.

Results

Relation between plasma phenobarbitone level and drug dose

For the three drugs studied the calculated coefficients of determination (see Figure 1) suggested that one or other form of curvilinear regression fitted the relation between plasma phenobarbitone level and drug dose (on a body weight basis) marginally better than did simple linear regression. However, the improvement in fit was so small that rectilinear regression was used for all subsequent analysis of data. The linear regressions for plasma phenobarbitone level on drug dose are shown for the various drugs in Figure 1. The drug doses necessary to produce a steady state plasma phenobarbitone level of 15 μg/ml (a reasonable 'therapeutic' value) are 1.75 mg kg^{-1} day^{-1} of phenobarbitone, 2.75 mg kg^{-1} day^{-1} of methylphenobarbitone and 7.75 mg kg^{-1} day^{-1} of primidone.

Effect of age

Phenobarbitone. It was possible to define four age groups (0–4, 5–14, 15–40, and over 40 years) which showed statistically significant differences ($P < 0.05$) from their neighbours in either elevation, or slope, or both, for the regression lines for plasma phenobarbitone levels on phenobarbitone dose. The phenobarbitone dose required to produce a given plasma phenobarbitone level tended to fall progressively with age.

Methylphenobarbitone. For patients taking methylphenobarbitone, the techniques used permitted definition of three age groups (0–14 years; 15–40 years and over 40 years) which behaved differently ($P < 0.05$) from their neighbours in relation to the regression for plasma phenobarbitone level on methylphenobarbitone dose. Again, dosage requirement to produce a given phenobarbitone level fell with increasing age.

Primidone. For primidone it was not possible to determine age groups which differed to a statistically significant extent in relation to the regressions for plasma phenobarbitone level on drug dose.

The various regressions for the three drugs are shown in Figure 2.

Effect of sex

Phenobarbitone. For patients taking phenobarbitone itself, the regressions for plasma drug level on drug dose differed between males and females (Figure 3). Males tended to require higher drug doses

Table 1 Numbers of patients taking commonly-used non-barbiturate anticonvulsants

	Number taking	*Number taking*		
		phenytoin	*carbamazepine*	*sulthiame*
Phenobarbitone	121	69	25	3
Methylphenobarbitone	123	89	21	12
Primidone	58	52	13	0

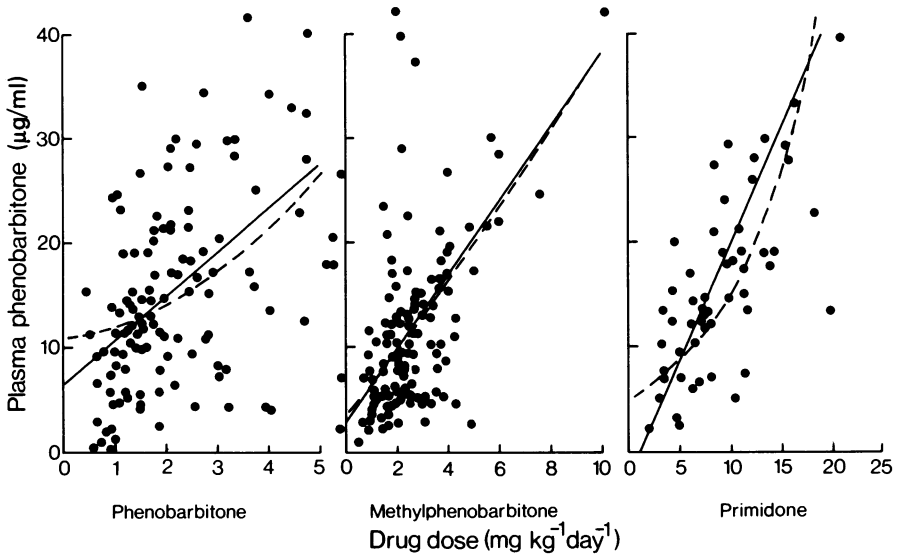

Figure 1 Plasma phenobarbitone levels plotted against doses of phenobarbitone, methylphenobarbitone and primidone in the patients studied. For phenobarbitone, the regression for the curve of best fit was $y = 11.147 + 0.440x + 0.521x^2$ ($r^2 = 0.331$); for methylphenobarbitone $y = 3.696 + 3.071x + 0.037x^2$ ($r^2 = 0.432$); and for primidone $y = 5.239e^{.109x}$ ($r^2 = 0.495$).

The corresponding linear regressions were, respectively, $y = 6.452 + 4.226x$($r^2 = 0.312$); $y = 2.902 + 3.513x$ ($r^2 = 0.430$); and $y = -2.628 + 2.278x$ ($r^2 = 0.452$).

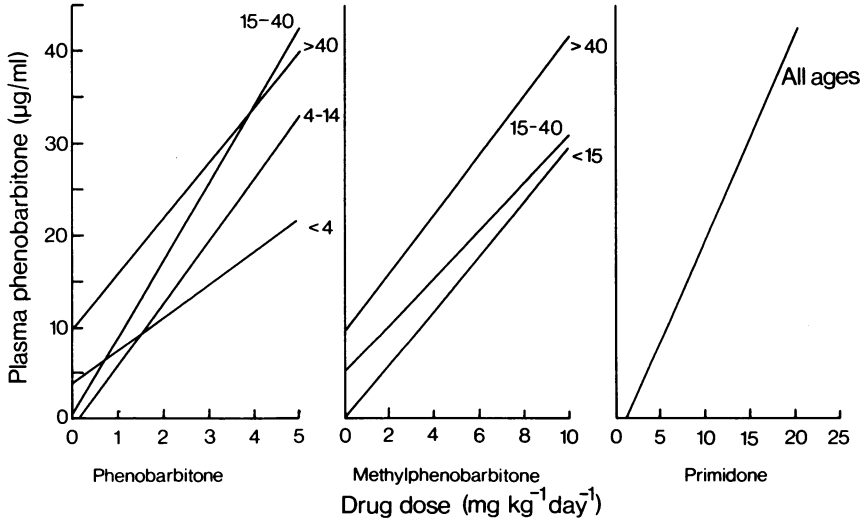

Figure 2 Linear regressions for plasma phenobarbitone level on drug dose in subjects in different age groups. The regressions shown are those which differed to a statistically significant extent ($P < 0.05$).

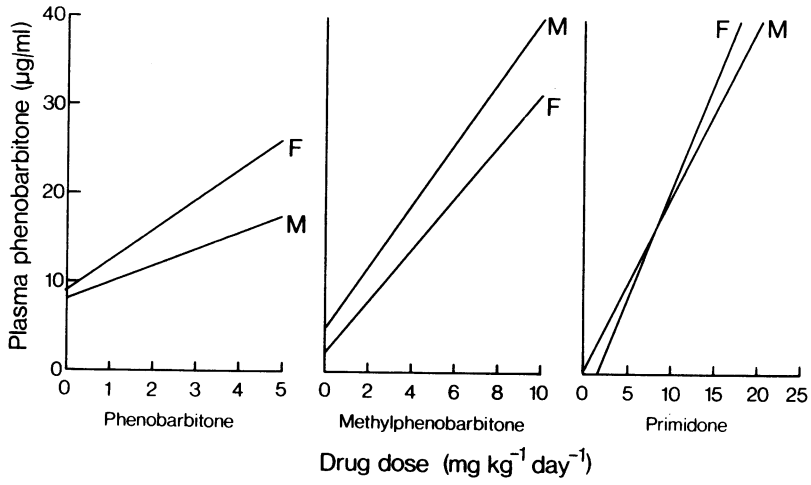

Figure 3 Linear regressions for plasma phenobarbitone level on drug dose in male and female subjects.

than females to achieve the same plasma phenobarbitone levels. The effect of sex on the relation between plasma drug level and drug dose was studied in the various age groups which had differed in their phenobarbitone dose requirements. It was found that sex affected the relationship between plasma level and drug dose only in children below 5 years, in whom males had a higher dosage requirement than females. In these young children there was no statistically significant difference in the mean phenobarbitone dose taken by males (3.67 mg kg^{-1} day^{-1}) and by females (3.98 mg kg^{-1} day^{-1}) ($t=0.529$; d.f. $=34$; $P>0.5$).

Methylphenobarbitone. The regression for plasma phenobarbitone level on methylphenobarbitone dose also differed between the sexes (Figure 3). In contradistinction to the situation applying for phenobarbitone, males tended to require lower methylphenobarbitone doses than females to produce the same plasma phenobarbitone level. The regressions were virtually identical for males and females under 14 years, but differed to a statistically significant extent between the sexes in both of the older age groups studied.

Primidone. There were no statistically significant difference between the sexes for the slopes and elevations of the regressions for plasma phenobarbitone level on primidone dose.

Table 2 Multiple linear regression analysis of the relation between plasma phenobarbitone level (y) in μg/ml and doses of phenobarbitone (or methylphenobarbitone or primidone) (x_1), phenytoin (x_2), carbamazepine (x_3) and sulthiame (x_4), expressed in mg kg^{-1} day^{-1}. The regression equation has the form

$$y=a+bx_1+cx_2+dx_3+ex_4.$$

Corresponding partial correlation coefficients are shown.

For phenobarbitone $y=10.766+6.408x_1-1.396x_2-0.128x_3+0.138x_4$				
and corresponding r (partial correlation coefficients) are	=0.682*	0.054	0.117	0.093
For methylphenobarbitone $y=9.574+3.066x_1-0.759x_2-0.281x_3-0.205x_4$				
	r=0.580*	0.105	0.090	0.153
For primidone† $y=1.854+1.748x_1+0.259x_2+0.921x_3$				
	r=0.649*	0.379*	0.558*	

* $P<0.05$.

† No patients taking primidone were receiving sulthiame.

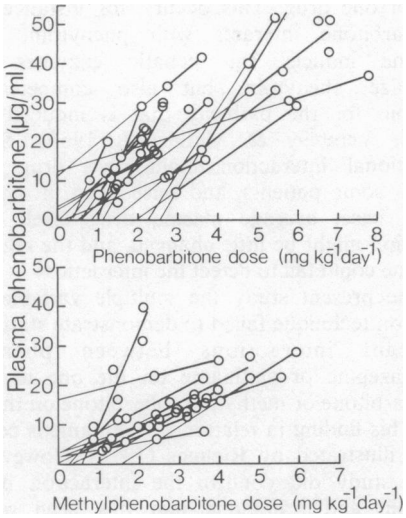

Figure 4 Individual patients' regressions for steady state plasma phenobarbitone levels on drug dose extrapolated back toward zero.

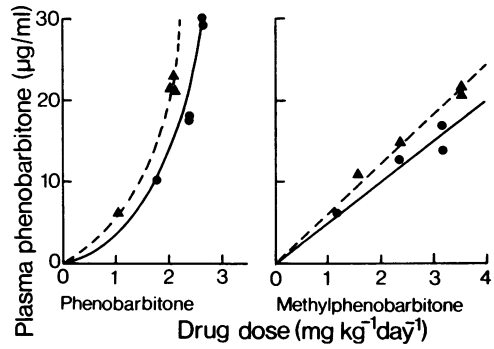

Figure 5 (a) The curvilinear relation between steady state plasma phenobarbitone level and phenobarbitone dose in two subjects, and (b) the rectilinear relation between plasma phenobarbitone level and methylphenobarbitone dose in another two subjects.

Effects of other anticonvulsants

Phenobarbitone. Multiple variable linear regression analysis detected no statistically significant effect of phenytoin, carbamazepine or sulthiame dosage on the relation between plasma phenobarbitone level and phenobarbitone dose (Table 2).

Methylphenobarbitone. Again, multiple variable linear regression analysis detected no statistically significant effect of phenytoin, carbamazepine or sulthiame dosage on the regression for plasma phenobarbitone level on methylphenobarbitone dose (Table 2).

Primidone. Dosage of phenytoin, and of carbamazepine, had statistically significant effects in increasing the amount of phenobarbitone present in plasma in patients taking primidone (Table 2).

Effects of dosage increments in the individual

Phenobarbitone. Steady state plasma phenobarbitone levels in individual patients who had taken different phenobarbitone doses at different times were plotted and the regression lines joining values for individuals were extrapolated backwards towards the origin (Figure 4). Thirteen of seventeen lines intersected the x (dosage) axis and four the y (plasma level) axis. This is a statistically significant difference ($P=0.05$: 'exact' test). While there are several possible explanations for this finding, the data of Figure 5 suggest that the relation between plasma phenobarbitone level and phenobarbitone dose in the individual may be curvilinear. This could explain the findings of Figure 4.

Methylphenobarbitone. Unlike the situation for phenobarbitone, the extrapolated individual regression lines for patients taking different methylphenobarbitone doses showed an evenly distributed scatter of intercepts around the origin, seven lines intersecting the x axis, seven the y axis (Figure 4). For methylphenobarbitone the relation between plasma level and drug dose in the individual appeared to be rectilinear (Figure 5).

Primidone. Insufficient data were available for analysis.

Discussion

The extent of the scatter of individual data points about the population regression lines for steady state plasma phenobarbitone level on dose of phenobarbitone, methylphenobarbitone or primidone is comparable to that found for other anticonvulsants, e.g. phenytoin (Eadie & Tyrer, 1974). In part this scatter represents the effects of biological variation, though incorrect compliance with prescribed drug dosage may also contribute. It is instructive to compare the doses of phenobarbitone, methylphenobarbitone and primidone necessary to produce a plasma phenobarbitone level of 15 μg/ml in the average patient (weighing 70 kg). Such a plasma phenobarbitone level is likely to be associated with a

reasonable therapeutic anticonvulsant effect in patients with appropriate types of epilepsy. The required dose (calculated from the data of Figure 1) is approximately 120 mg/day for phenobarbitone, 190 mg/day for methylphenobarbitone, and 540 mg/day for primidone. However, for patients taking primidone alone, the dose would be 675 mg (calculated from the multiple variable regression shown in Table 2). The lower dose calculated from Figure 1 is due to the effects of interactions. Relating these calculated values to the widely used adult doses of 30, 60 and 250 mg three times daily for the three drugs respectively, it is clear that from the point of view of producing desired plasma phenobarbitone levels the conventional dosage units of the drugs tend to favour primidone over the other drugs, and methylphenobarbitone over phenobarbitone.

In the present studies data were not available for neonates, but for all other ages there was a tendency for phenobarbitone and methylphenobarbitone dosage requirement (expressed on a body weight basis) to diminish with age. This effect was not detected in patients taking primidone. However there were fewer patients taking this drug and most of these patients were adults. Hence there may not have been sufficient younger patients to permit detection of an effect of age on dose requirement, if such an effect existed. An effect of age on dose requirement has been found for another anticonvulsant, phenytoin (Eadie, Tyrer & Hooper, 1973).

However, for phenytoin the relation between plasma drug level and dose, changes about the age of puberty and appears reasonably stable above and below this age. The factors involved in the progressive change in phenobarbitone and methylphenobarbitone dose requirement with age should be studied further.

Sex did not have a statistically significant effect on the relation between plasma drug level and dose in the case of phenytoin (Eadie *et al.*, 1973), or carbamazepine (Hooper, Dubetz, Eadie & Tyrer, 1974). In the present study sex had no effect on plasma phenobarbitone levels in patients taking primidone. The effect of sex in altering the relation between plasma phenobarbitone levels and phenobarbitone dose in the present study appeared, surprisingly, to be statistically significant ($P < 0.01$) only in children under 5 years. It might be wise to see if this finding is reproduced in another series of cases before it is accepted as valid. In the case of methylphenobarbitone sex again altered the relation between plasma phenobarbitone level and drug dose, but here the effect occurred in the age groups where it might have seemed more likely viz. persons over 14 years, rather than in young children.

The multiple variable linear regression technique used in the present study for detecting interactions has certain limitations. For instance, an interaction between two drugs might involve two mechanisms, one tending to raise and the other to lower the plasma levels of one drug. This occurs, for instance, when phenobarbitone interacts with phenytoin. Phenobarbitone induces the hepatic enzymes which metabolize phenytoin, but also competes with phenytoin for the pathway that is induced (Kutt, Haynes, Verebely & McDowell, 1969). Such a 'bidirectional' interaction might lower drug plasma levels in some patients, and raise them in others. In such a case, average plasma drug levels in the population might be little changed, and the regression technique could fail to detect the interaction.

In the present study the multiple variable linear regression technique failed to demonstrate statistically significant interactions between phenytoin, carbamazepine or sulthiame on the one hand and phenobarbitone or methylphenobarbitone on the other hand. This finding in relation to sulthiame is contrary to that illustrated by Richens (1976). However, the present study did confirm the interaction between phenytoin and primidone that has been reported previously by Reynolds, Fenton, Fenwick, Johnson & Laundy (1975). This interaction led to raised plasma phenobarbitone levels in patients taking primidone with phenytoin. The present regression analysis suggested that the typical adult phenytoin dose of 5 mg per kg per day would raise plasma phenobarbitone levels by some 1.3 μg/ml in the average patient taking primidone. The multivariate regression analysis also suggested the existence of a hitherto unreported interaction in which carbamazepine therapy raises plasma phenobarbitone levels in patients taking primidone. The commonly used carbamazepine dose of 10 mg/kg/day would tend to raise the plasma phenobarbitone level in the average patient taking primidone by about 9.0 μg/ml.

The evidence obtained in the present study suggests that the relation between plasma phenobarbitone level and phenobarbitone dose in the individual is not a straight line one, contrary to what has been stated previously (Richens, 1974). However, the relation between phenobarbitone level in plasma and methylphenobarbitone dose does appear rectilinear. Therefore the curvilinear relation in the case of phenobarbitone itself is unlikely to be due to poor assay techniques, since the same methods were used to measure plasma phenobarbitone concentrations in patients taking methylphenobarbitone. There are several possible explanations for the curvilinear relation between plasma phenobarbitone levels and phenobarbitone dose in the individual. For instance an elimination mechanism, probably biotransformation, might approach saturation as dose increased, or the apparent volume of distribution of the drug might decrease as dose increases. Wilson & Wilkinson (1973) did obtain some evidence of capacity-limited elimination of phenobarbitone in one subject. Further work is required to determine the mechanism involved in the absence of a straight line relationship between plasma level of phenobarbitone and its dose, but the

finding needs to be kept in mind in adjusting phenobarbitone doses in patients. Further work is also required to determine why the shape of the relation between plasma phenobarbitone level and drug doses differs when phenobarbitone is supplied directly, or supplied indirectly, by metabolism of methylphenobarbitone. In a personal, as yet unpublished, study of one subject, the data obtained were consistent with the possibility that phenobarbitone derived from methylphenobarbitone is eliminated in a quantitatively different fashion from phenobarbitone given as such.

The data provided by the present studies may be used to determine more appropriate doses of phenobarbitone (or of its congeners) in epileptic patients. In some ways the therapeutic situation is easiest to interpret when phenobarbitone itself is used, because only a single pharmacologically active substance is thought to be present in the body. However, the absence of a straight line relationship between plasma level and dose in the individual is a disadvantage in adjusting dosage when phenobarbitone itself is used. This disadvantage does not appear to apply for methylphenobarbitone over its usual dosage range. The possibility of interactions with other commonly used anticonvulsants adds an element of uncertainty to the situation when primidone is used. Therefore, if a barbiturate anticonvulsant or primidone is to be used, and facilities for plasma phenobarbitone measurement are not available, it may be easiest to prescribe appropriate drug doses if methylphenobarbitone is the drug chosen.

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