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ELECTROLYTE COMPOSITION OF PAROTID SALIVA FROM SODIUM-REPLETE RED KANGAROOS (*MACROPUS RUFUS*)

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SUMMARY

Saliva was collected from the parotid salivary gland of anaesthetized sodium-replete red kangaroos (*Macropus rufus*) by catheterization of the parotid duct through its opening in the mouth. Salivary secretion was stimulated by ipsilateral intracarotid infusion of acetylcholine at varying rates to produce salivary flow rates ranging from $0{\cdot}056 \pm 0{\cdot}0042$ (S.E. of mean) to $4{\cdot}509 \pm 0{\cdot}1136\,\mathrm{ml\,min^{-1}}$. The concentrations of sodium ($142{\cdot}2 \pm 1{\cdot}93$ to $157{\cdot}0 \pm 1{\cdot}17\,\mathrm{mmol\,l^{-1}}$), calcium ($40{\cdot}1 \pm 7{\cdot}08$ to $72{\cdot}8 \pm 8{\cdot}0\,\mu\mathrm{mol\,l^{-1}}$) and bicarbonate ($68{\cdot}6 \pm 3{\cdot}48$ to $143{\cdot}3 \pm 0{\cdot}67\,\mathrm{mmol\,l^{-1}}$) and the osmolality ($270{\cdot}1 \pm 2{\cdot}98$ to $291{\cdot}7 \pm 2{\cdot}10\,\mathrm{mosmol\,kg^{-1}}$) were positively correlated with salivary flow rate, whereas the concentrations of potassium ($11{\cdot}4 \pm 0{\cdot}57$ to $6{\cdot}92 \pm 0{\cdot}19\,\mathrm{mmol\,l^{-1}}$), magnesium ($206{\cdot}0 \pm 34{\cdot}1$ to $9{\cdot}3 \pm 0{\cdot}78\,\mu\mathrm{mol\,l^{-1}}$), hydrogen ion ($17{\cdot}0 \pm 1{\cdot}89$ to $6{\cdot}82 \pm 0{\cdot}49\,\mathrm{nmol\,l^{-1}}$), chloride ($30{\cdot}7 \pm 2{\cdot}41$ to $4{\cdot}11 \pm 0{\cdot}23\,\mathrm{mmol\,l^{-1}}$) and phosphate ($47{\cdot}6 \pm 2{\cdot}65$ to $14{\cdot}9 \pm 0{\cdot}81\,\mathrm{mmol\,l^{-1}}$) were negatively correlated with flow rate. The relationships between flow rate and concentration were curvilinear for all the inorganic solutes. The rates of secretion for each ion and for total solute were positively correlated with salivary flow rate. These regressions for sodium, potassium, calcium, hydrogen ion, bicarbonate and osmolality were always linear, with highly significant correlation coefficients and variance ratios, which indicated that the changes in concentration of these ions were related solely to flow rate and were not due to any other factor modifying glandular function. Spontaneous secretion was not observed during anaesthesia.

INTRODUCTION

Characteristically, ruminants secrete large volumes of parotid saliva, which has high levels of sodium and high buffering capacity due to the presence of bicarbonate and phosphate in relatively large amounts. A major function of this saliva is to prevent a fall in pH in the foregut which would otherwise occur as an end-product of the fermentation in this chamber. The same problem must be solved by non-ruminant foregut fermenters, but little is known of the possible role of saliva in this function since salivary composition and secretion rates have rarely been investigated in these

Key words: Electrolytes, parotid saliva, red kangaroo.

animals. A small amount of data exists for camels and this indicates that their parotid saliva composition is very similar to that of ruminants (Hoppe, Kay & Maloiy, 1975). However, camels are very close to ruminants in evolutionary terms (some authorities have incorporated the Tylopoda into the Ruminantia) whereas marsupials are not. Partial analyses of the composition of parotid saliva have been reported for one sample from a conscious euro (*Macropus robustus*) and for one sample from an anaesthetized red kangaroo (Brown, 1964; Forbes & Tribe, 1969). Both samples had high sodium concentrations. Unfortunately, the salivary flow rate and sodium status of the animals were not given in either report and the blood supply to the parotid gland of the red kangaroo may have been damaged.

This paper reports a comprehensive investigation of the inorganic composition of parotid saliva from sodium-replete red kangaroos over a wide range of flow rates. In addition, the paper addresses the question of how the 'buffering' capacity of this saliva compares with parotid saliva of ruminants and the wider question of whether the type of parotid saliva, characteristic of ruminants, is a necessary adjunct to digestion by foregut fermentation.

METHODS

Experimental procedures

Twelve experiments were performed on six adult red kangaroos (four males weighing 33–41 kg and two non-lactating females weighing 22·5–25·5 kg). Each animal had one carotid artery exteriorized in a skin loop and was allowed a minimum of one month for recovery and healing before the animals underwent any experiments. The kangaroos were maintained on a diet of lucerne chaff, concentrate cubes and water *ad lib.*, the water being replaced with a dilute saline solution (25 mmol l^{-1} NaCl+25 mmol l^{-1} $NaHCO_3$) for periods ranging from 1–8 weeks prior to saliva collection.

Two days before each experiment the animals were lightly anaesthetized with Ketamine hydrochloride (Ketalar; Park Davis, Australia) and one lateral tail vein was cannulated with a vinyl cannula (0·86 mm i.d., 1·27 mm o.d.; Dural Plastics, N.S.W.) using the technique of Seldinger (1953). To ensure that the kangaroos were sodium-replete, each animal was given 60 ml of 2 mol l^{-1} NaCl solution by slow intravenous injection *via* this cannula. The cannula was then filled with heparinized saline (1000 i.u. ml^{-1}) and protected with a bandage. Food was removed 15–16 h before commencement of each experiment while the saline drinking solution was available until the experiment began.

At the beginning of each experiment, the kangaroos were anaesthetized with 5 % sodium pentobarbitone in saline given by i.v. injection through the tail vein cannula. Anaesthesia was maintained with sodium pentobarbitone throughout the experiment. The animals were positioned on one side (carotid loop side up) with a heated pad under the thorax to maintain normal body temperature and with an air cushion under the hind quarters to prevent pressure damage to the hip and thigh region. The trachea was intubated with an endotracheal tube which was shortened so that respiratory dead space was not increased. A solution of NaCl: KCl (150: 4 mmol l^{-1}) was infused intravenously at 1·4–2·0 ml min^{-1} for the duration of each experiment to minimize changes in body fluid composition resulting from transpiration and salivary loss. The

Parotid artery loop was cannulated with a polyethylene cannula (0·58 mm i.d., 0·96 mm o.d.; Dural Plastics, N.S.W.) inserted 10 cm in the direction of the heart using the technique of Seldinger (1953). The duct of the parotid gland ipsilateral to the carotid artery loop was catheterized with a vinyl tube (1·40 mm i.d., 1·90 mm o.d. or 1·57 mm i.d., 2·08 mm o.d.; Dural Plastics, N.S.W.) introduced 3 cm into the duct through its orifice in the mouth. Saliva was collected into polystyrene and polypropylene sample tubes which were closed except for a 20 wire gauge air-bleed. The distal end of the salivary catheter was about 10 cm below the duct orifice and the dead space in the catheter was 0·4–0·5 ml.

Salivary secretion was stimulated by intracarotid infusion of acetylcholine chloride (Sigma Chemical Co., U.S.A.) at rates ranging from 4·5–500 nmol min^{-1} using a variable speed syringe pump. Each kangaroo underwent two experiments which were not less than 1 month apart. In the first experiment, salivary secretion was stimulated to produce a low flow rate (<0·125 ml min^{-1}) with secretion being increased subsequently to maximum through a series of predetermined flow rate ranges by increasing the rate of acetylcholine infusion. The flow rate ranges used were <0·125, 0·125–0·25, 0·25–0·5, 0·5–1·0, 1·0–2·0, 2·0–3·0, 3·0–4·0 and >4·0 ml min^{-1}. In the second experiment, maximal flow was stimulated initially and thereafter, by reducing the rate of acetylcholine infusion, a series of decreasing flow rates falling within the same flow intervals were obtained. During any period of stimulation, salivary collection was not commenced until the salivary flow was well established and reasonably constant. Two or more timed samples were taken during every flow interval (except <0·125 ml min^{-1} interval) and the results obtained from their analysis were averaged to provide mean values for each flow interval of each experiment. Rest periods of 10–60 min were allowed between periods of gland stimulation, the shortest rest periods followed the period of lowest stimulation and the longest rest periods followed maximal stimulation. Blood samples (6–8 ml) were taken from the carotid artery before the first acetylcholine infusion and thereafter were taken at the end of each period of gland stimulation. Blood plasma and saliva were analysed for sodium, potassium, calcium, magnesium, chloride, phosphate and osmolality, and additionally saliva was analysed for hydrogen ion and bicarbonate.

In three additional experiments, three of the kangaroos were pretreated over the 24 h before experiment with spironolactone (Aldactone, Searle & Co.) at a rate of 2 mg kg^{-1}. The spironolactone was dissolved in ethyl oleate and was given intramuscularly in two doses. Salivation was stimulated by intracarotid acetylcholine infusion to produce salivary flow rates in the 0·25–0·5 and the 2–3 ml min^{-1} ranges. Only plasma and saliva sodium and potassium concentrations were estimated in these experiments.

Analytical procedures

Blood samples were taken into plastic syringes heparinized with one drop of heparin (5000 i.u. ml^{-1}) and centrifuged at 2200 ***g*** for 10 min to obtain plasma for analysis. Microhaematocrit determinations were made in triplicate on blood spun at 12 000 ***g*** for 10 min in a microhaematocrit centrifuge (Hawksley). Salivary pH was measured at 36 °C under anaerobic conditions using thermostatted Radiometer microelectrodes. Saliva and plasma were analysed in duplicate for sodium, potassium, calcium and

magnesium by atomic absorption spectroscopy using standards containing appropriate ionization suppressants. Duplicate estimations of the chloride concentration in plasma and saliva were made using a Radiometer chloride titrator (model CMT 10). Total inorganic phosphate concentrations in saliva and plasma were determined in duplicate using the method of Baginski, Foa & Zak (1967). The osmolality of plasma and saliva was estimated by freezing point depression using a Knauer osmometer. The bicarbonate concentration of saliva was determined by the titration procedure of Gyory & Edwards (1967) modified for 0·2 ml aliquots of sample.

Statistical procedures

The data for salivary and plasma electrolyte concentrations were analysed by one-way analysis of variance across the eight predetermined flow intervals. The rates of salivary electrolyte and osmolal secretion were regressed on salivary flow rate for each individual experiment and the regression coefficients, correlation coefficients and variance ratios of these regressions were calculated.

RESULTS

In all experiments, salivary flow from the parotid gland ceased once the kangaroo was anaesthetized, thus making stimulation of the gland essential for obtaining any saliva. In two experiments on each kangaroo, the rate of parotid saliva secretion was altered through eight different flow intervals either by increasing the flow from $<0{\cdot}125\ \text{ml min}^{-1}$ to the maximum sustainable flow rate or by decreasing flow from maximum to $<0{\cdot}125\ \text{ml min}^{-1}$. As no significant differences were found in the electrolyte concentrations of saliva from the same flow interval of the two experimental regimes, the data for the experiments were combined. Mean values for the haematocrit, plasma osmolality and the plasma concentrations of sodium, potassium, calcium, magnesium, chloride and phosphate from carotid arterial blood were estimated for each of the eight flow rate intervals (Figs 1–4). No significant differences were found between the flow rate intervals in haematocrit, plasma osmolality or plasma electrolyte concentrations.

The sodium concentration of the saliva was positively correlated with saliva flow rate and exceeded the plasma sodium concentration at all but the lowest flow rate (Fig. 1). At the lowest flow rate ($0{\cdot}056 \pm 0{\cdot}0042\ \text{ml min}^{-1}$), the salivary sodium concentration ($142{\cdot}2 \pm 1{\cdot}93\ \text{mmol l}^{-1}$) was significantly lower ($t_{10} = 2{\cdot}93$; $P < 0{\cdot}02$) than that of the corresponding plasma concentration ($147{\cdot}1 \pm 1{\cdot}01\ \text{mmol l}^{-1}$). The rate of secretion of sodium in the saliva rose from $7{\cdot}9 \pm 0{\cdot}65\ \mu\text{mol min}^{-1}$ at the lowest flow rate to $655{\cdot}7 \pm 31{\cdot}03\ \mu\text{mol min}^{-1}$ at the maximal flow rate for all experiments ($4{\cdot}18 \pm 0{\cdot}190\ \text{ml min}^{-1}$). For each experiment this regression was highly linear (variance ratios for the regressions ranged from 19 138–2 608 664) which made feasible the estimation of the maximum sodium concentration which would occur under

Fig. 1. Salivary sodium and potassium concentrations from the parotid gland of red kangaroos regressed on salivary flow rate. Sodium and potassium concentrations of arterial plasma are given for each flow rate ($N = 12$; means ± S.E. of mean). Only three of the kangaroos contributed to the highest flow rate (open circles).

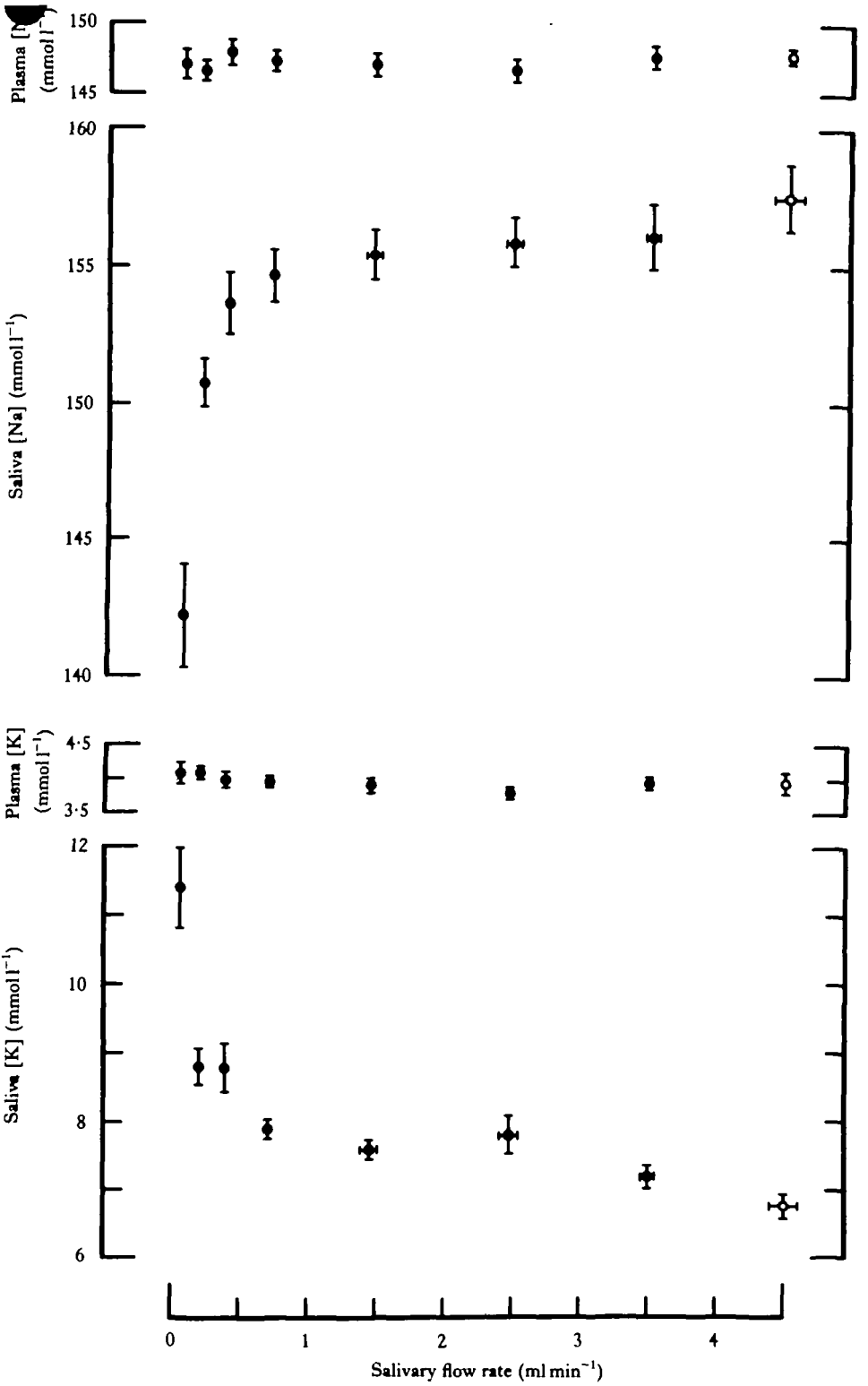
Plasma [Na] (mmol l⁻¹)
150
145
Saliva [Na] (mmol l⁻¹)
160
155
150
145
140
Plasma [K] (mmol l⁻¹)
4·5
3·5
Saliva [K] (mmol l⁻¹)
12
10
8
6
0
1
2
3
4
Salivary flow rate (ml min⁻¹)

Fig. 1

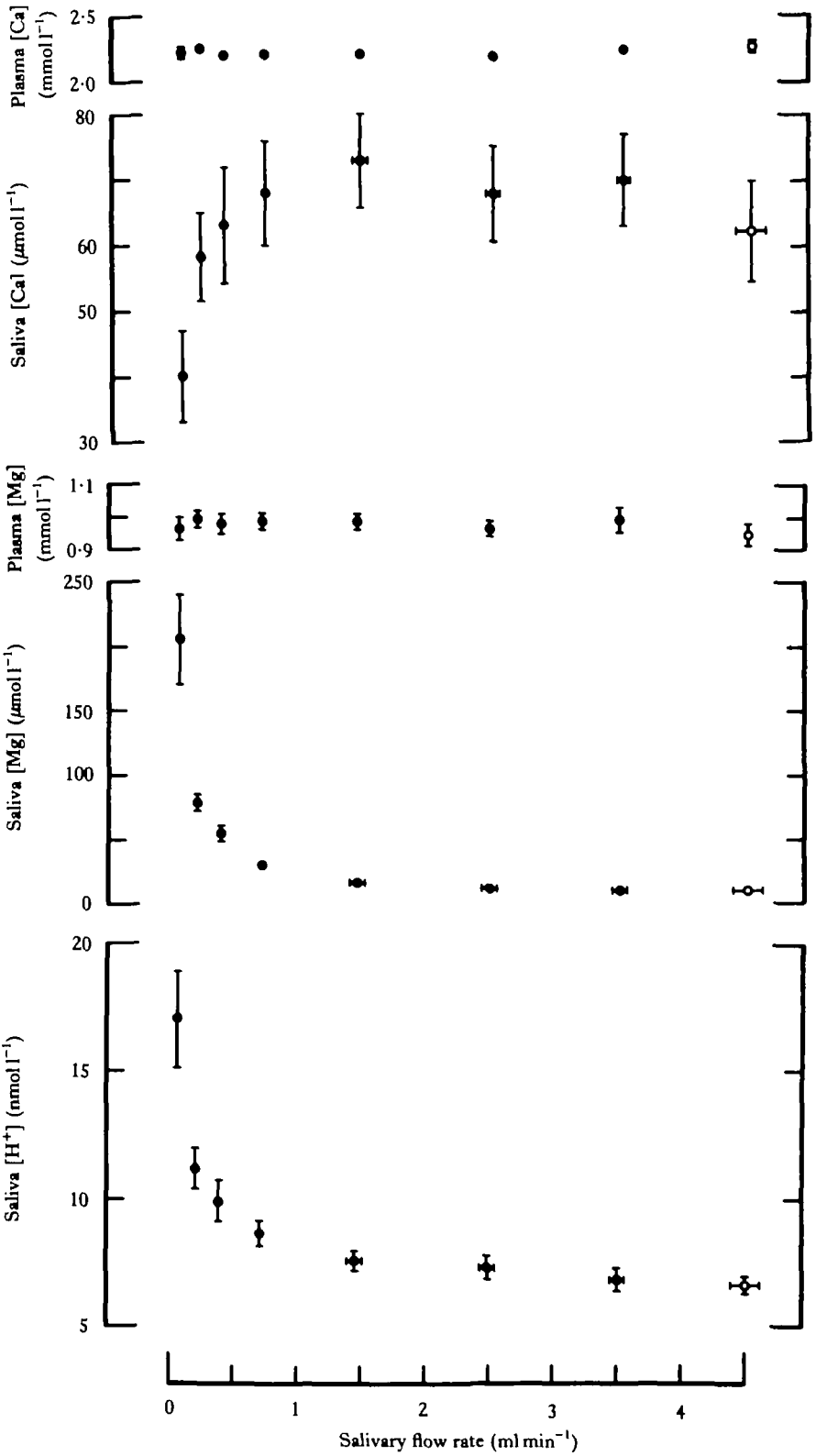
Plasma [Ca] (mmol l⁻¹)
2·5
2·0
Saliva [Ca] (μmol l⁻¹)
80
60
50
30
Plasma [Mg] (mmol l⁻¹)
1·1
0·9
Saliva [Mg] (μmol l⁻¹)
250
150
100
0
Saliva [H⁺] (nmol l⁻¹)
20
15
10
5
0
1
2
3
4
Salivary flow rate (ml min⁻¹)

Fig. 2

Table 1. *Salivary flow rates and the concentrations of sodium and potassium in parotid saliva from red kangaroos pretreated with spironolactone*

Flow rate ($ml\ min^{-1}$)	Sodium concentration ($mmol\ l^{-1}$)	Potassium concentration ($mmol\ l^{-1}$)
$0{\cdot}42 \pm 0{\cdot}027$	$154{\cdot}7 \pm 0{\cdot}40$	$8{\cdot}29 \pm 0{\cdot}277$
$2{\cdot}56 \pm 0{\cdot}211$	$156{\cdot}6 \pm 0{\cdot}15$	$6{\cdot}78 \pm 0{\cdot}419$

$N = 3$; means ± S.E. of mean.

these experimental conditions, i.e. the sodium concentration at infinite flow ($157{\cdot}5 \pm 0{\cdot}94\ mmol\ l^{-1}$). The salivary potassium concentration exceeded the plasma potassium concentration at all flow rates and was negatively correlated with salivary flow (Fig. 1). At the same time, the rate of salivary potassium secretion increased linearly from $0{\cdot}62 \pm 0{\cdot}034\ \mu mol\ min^{-1}$ at the lowest flow rate to $28{\cdot}71 \pm 1{\cdot}107\ \mu mol\ min^{-1}$ at maximum flow. The variance ratios of the secretion rate regressions ranged from 539–6078 and the estimated asymptote for salivary potassium concentration was $7{\cdot}07 \pm 0{\cdot}180\ mmol\ l^{-1}$. The concentrations of sodium and potassium in the saliva from kangaroos pretreated with spironolactone were within the range of values obtained for saliva of similar flow rate from untreated animals (Table 1).

The concentration of calcium in the saliva was positively correlated with flow rate. The concentration was very low, the mean concentration being approximately $63\ \mu mol\ l^{-1}$ or 0·03 % of the plasma calcium concentration (Fig. 2). Salivary calcium secretion rate increased from $2{\cdot}32 \pm 0{\cdot}492\ nmol\ min^{-1}$ at low flow rates to $267{\cdot}7 \pm 26{\cdot}00\ nmol\ min^{-1}$ at maximal flow. The secretion rate regressions were linear (variance ratios of 624–19 561) and the estimated maximum concentration of calcium in the saliva at infinite flow was $72{\cdot}8 \pm 8{\cdot}27\ \mu mol\ l^{-1}$. Salivary magnesium concentration was negatively correlated with flow rate (Fig. 2) and varied between 21 and 1 % of the plasma magnesium concentration. The rate of magnesium secretion increased with variable linearity from $11{\cdot}1 \pm 1{\cdot}49\ nmol\ min^{-1}$ at the lowest flow rate to $39{\cdot}3 \pm 4{\cdot}22\ nmol\ min^{-1}$ at maximum flow rates.

The mean hydrogen ion concentration of the saliva decreased from a maximum of $17{\cdot}0 \pm 1{\cdot}89\ nmol\ l^{-1}$ (pH = 7·77) at the lowest flow rate to $6{\cdot}9 \pm 0{\cdot}51\ nmol\ l^{-1}$ (pH = 8·16) at the mean maximum flow rate (Fig. 2). The rate of hydrogen ion secretion increased linearly from a mean value of $0{\cdot}94 \pm 0{\cdot}109\ pmol\ min^{-1}$ at the

Fig. 2. Salivary calcium, magnesium and hydrogen ion concentrations from the parotid gland of red kangaroos regressed on salivary flow rate. Calcium and magnesium concentrations of arterial plasma are given for each flow rate ($N = 12$; means ± S.E. of mean). Only three of the kangaroos contributed to the highest flow rate (open circles).

Fig. 3. Salivary bicarbonate and inorganic phosphate concentrations from the parotid gland of red kangaroos regressed on salivary flow rate. Inorganic phosphate concentrations of arterial plasma are given for each flow rate ($N = 12$; means ± S.E. of mean). Only three of the kangaroos contributed to the highest flow rate (open circles).

Fig. 4. Salivary chloride and osmolal concentrations from the parotid gland of red kangaroos regressed on salivary flow rate. Chloride and osmolal concentrations of arterial plasma and arterial blood haematocrit are given for each flow rate ($N = 12$; means ± S.E. of mean). Only three of the kangaroos contributed to the highest flow rate (open circles).

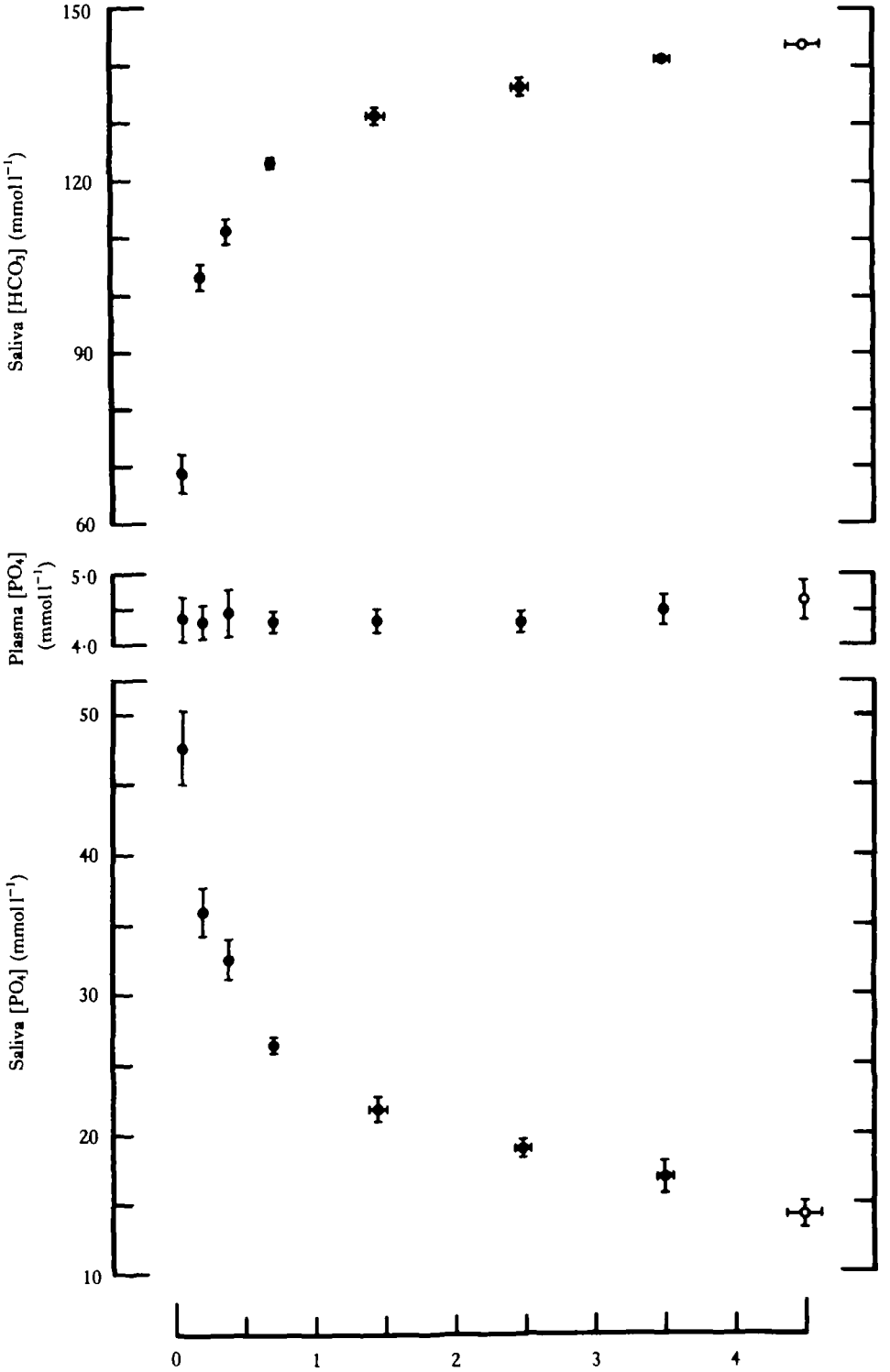

Fig. 3 for legend see p. 231

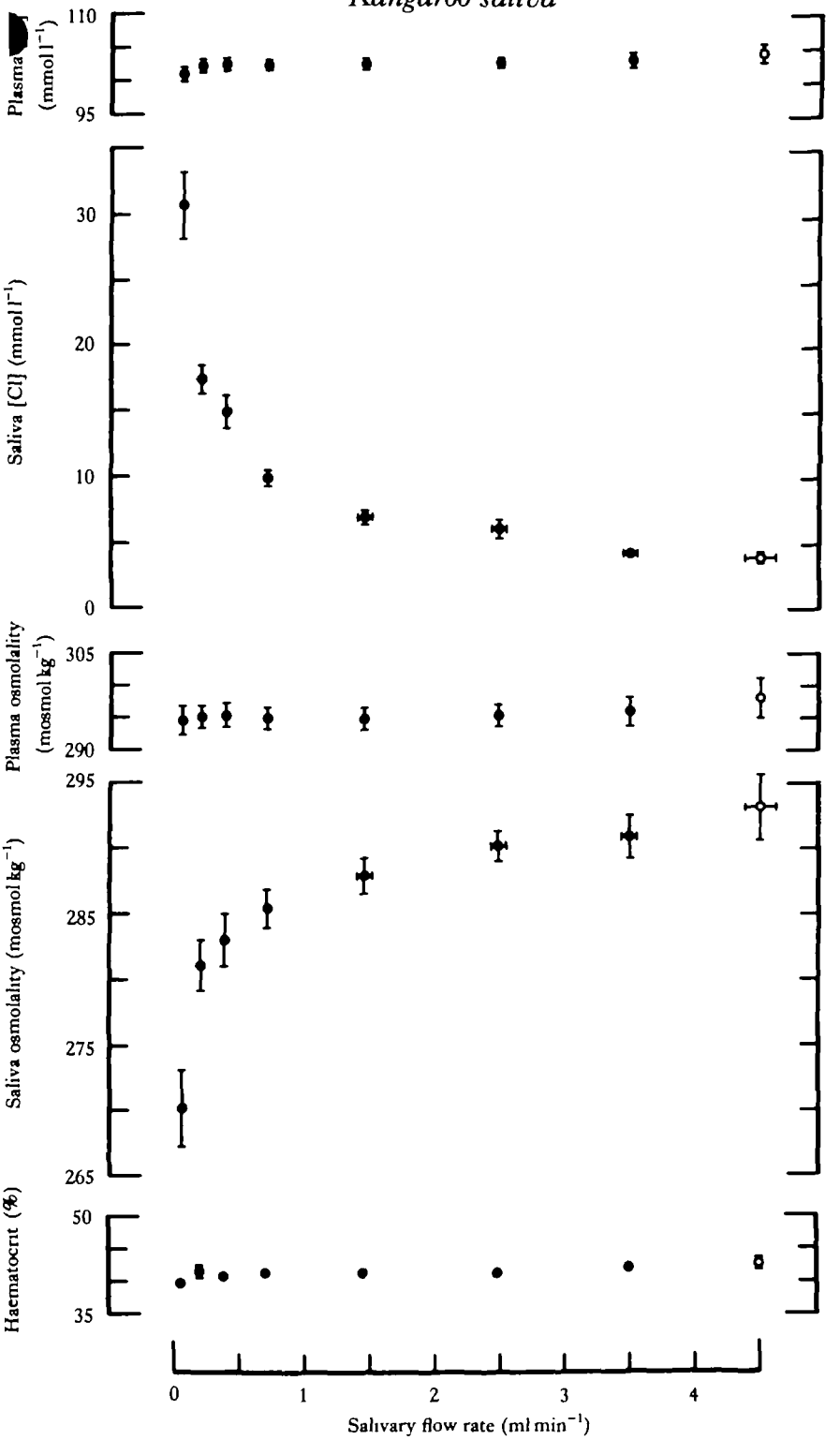

Fig. 4 for legend see p. 231

lowest flow rate to $28{\cdot}8 \pm 2{\cdot}18$ pmol min^{-1} at maximum flow rates. The variance ratios of the secretion rate regressions ranged from 166–7903 and hydrogen ion concentration was estimated to level off at $6{\cdot}43 \pm 0{\cdot}507$ nmol l^{-1}. Salivary bicarbonate concentration was positively correlated with salivary flow rate (Fig. 3), rising from $68{\cdot}6 \pm 3{\cdot}48$ to $143{\cdot}3 \pm 0{\cdot}67$ mmol l^{-1} at maximum flow. The rate of salivary secretion of bicarbonate increased from $3{\cdot}9 \pm 0{\cdot}43$ μmol min^{-1} at the lowest flow rate to $598{\cdot}7 \pm 28{\cdot}14$ μmol min^{-1} at maximal flow rates. The secretion rate of bicarbonate was linearly related to flow (variance ratios of 4455–24 176), and the maximum concentration at infinite flow was estimated to be $145{\cdot}9 \pm 1{\cdot}07$ mmol l^{-1}. The concentration of inorganic phosphate in the saliva fell with increasing salivary flow rate from $47{\cdot}6 \pm 2{\cdot}65$ mmol l^{-1} ($90{\cdot}4 \pm 0{\cdot}96$ % HPO_4^{2-}) at the lowest flow rates to $14{\cdot}9 \pm 0{\cdot}81$ mmol l^{-1} ($95{\cdot}8 \pm 0{\cdot}29$ % HPO_4^{2-}) at maximum flow rates (Fig. 3). Salivary phosphate secretion increased with variable linearity from $2{\cdot}63 \pm 0{\cdot}234$ μmol min^{-1} during the lowest flow interval at $61{\cdot}3 \pm 3{\cdot}13$ μmol min^{-1} at maximum flow rates.

The chloride concentration of the saliva was negatively correlated with saliva flow (Fig. 4), ranging from $30{\cdot}7 \pm 2{\cdot}41$ mmol l^{-1} at the low flow rate interval to $4{\cdot}1 \pm 0{\cdot}23$ mmol l^{-1} at maximum flow. The rate of salivary chloride secretion increased with variable linearity from $1{\cdot}67 \pm 0{\cdot}132$ μmol min^{-1} at low flows to $17{\cdot}1 \pm 1{\cdot}24$ μmol min^{-1} at maximum flow. The saliva was slightly hyposmotic to plasma (92·1–98·7 %) at all flow rates, with the mean osmolality at the lowest flow interval being $270{\cdot}1 \pm 2{\cdot}98$ mosmol kg^{-1} increasing to $291{\cdot}7 \pm 2{\cdot}10$ mosmol kg^{-1} at maximum flow. At the same time, osmolal secretion rate increased from $15{\cdot}1 \pm 1{\cdot}19$ to $1219{\cdot}4 \pm 59{\cdot}06$ μosmol min^{-1}. The osmolal secretion rate regressions were linear (variance ratios of 33 910–1 783 763), giving an estimated maximum concentration at infinite flow of $291{\cdot}8 \pm 1{\cdot}43$ mosmol kg^{-1}.

DISCUSSION

The parotid gland of ruminants will secrete spontaneously at a low rate after being deprived of secretomotor stimulation by denervation of the gland or by barbiturate anaesthesia (Coats, Denton, Goding & Wright, 1956; Beal, 1977). Barbiturate anaesthesia abolished all flow from the kangaroo parotid gland thus demonstrating that the kangaroo gland, unlike that of ruminants but in common with most other salivary glands, does not secrete spontaneously. The maximum sustainable rate of saliva flow from the kangaroo parotid glands ($4{\cdot}18 \pm 0{\cdot}190$ ml min^{-1}) was higher than the mean sustainable maxima (2·6–3·0 ml min^{-1}) reported for the parotid glands of anaesthetized sheep (Coats & Wright, 1957; Coats, Denton & Wright, 1958; Beal, 1980). However, based on the body weights of the sheep (Beal, 1980) and kangaroos and the parotid gland weight/body weight ratios of 1·07 and 0·63 g gland/kg body weight for kangaroos and sheep respectively (Porter, 1981), the maximum flows from the parotid glands of sheep and kangaroos were very similar on a secretion rate/gland weight basis.

Characteristically, the parotid saliva of ruminants has a high pH associated with substantial concentrations of bicarbonate and alkaline phosphate and, as a result, has a high buffering capacity. The kangaroos were found to have concentrations of hydrogen ion, bicarbonate and phosphate in their parotid saliva which were simi

those found in the parotid saliva of sheep at comparable flow rates. The concentrations of hydrogen ion and phosphate in the saliva were negatively correlated in a curvilinear manner with salivary flow, as has been reported for sheep parotid saliva (McDougall, 1948; Denton, 1956; Coats & Wright, 1957; Kay, 1960; Beal, 1979). The concentration of bicarbonate in kangaroo parotid saliva was positively correlated with salivary flow rate in a curvilinear manner which has also been found for the parotid saliva of ruminants (McDougall, 1948; Coats & Wright, 1957; Kay, 1960; Beal, 1979) and commonly occurs in saliva secreted during parasympathetic stimulation of the salivary glands of various unrelated species. The equivalence of the concentrations of these ions in the parotid saliva of sheep and red kangaroos and their relation to salivary flow rate becomes even greater if the larger size of the kangaroo parotid gland is taken into consideration. Hence, the buffering capacity of red kangaroo parotid saliva is at least equal to that of sheep and other ruminants.

The concentration of sodium in kangaroo parotid saliva was positively correlated with salivary flow rate and exceeded the plasma sodium concentration at all but the very lowest flow rates. Salivary sodium concentrations in excess of the plasma concentration are relatively uncommon having been found in rats and ruminants such as sheep, goats and cattle (Schneyer & Hall, 1965; Coats & Wright, 1957; Beal, 1979; Komi & Snyder, 1963; Bailey & Balch, 1961). A positive correlation is the typical relation between saliva sodium concentration and flow rate for most salivas evoked by parasympathetic stimulation including that of sodium-deficient sheep (Denton, 1956; Beal, Clark & Budtz-Olsen, 1975). However, in 'sodium-replete' sheep, parotid salivary sodium concentration has been variously reported to be positively correlated with flow (Coats & Wright, 1957; Kay, 1960; Compton, Nelson, Wright & Young, 1980), to show no flow dependence (Compton *et al.* 1980) or to be negatively correlated with flow (Thaysen & Tarding, 1974; Kay, 1960; Beal, 1977, 1979, 1980). Histological evidence has been published which indicates that the parotid glands of kangaroos are responsive to low sodium status, presumably through the action of mineralocorticoids (Blair-West *et al.* 1968). Thus, given that it is not clear which sodium/flow relation represents the sodium-replete state in sheep and that the kangaroo parotid gland appears to respond to sodium depletion, it is important to try to establish the sodium status of the red kangaroos. There are a number of reasons for believing that the kangaroos were sodium-replete and had low mineralocorticoid levels throughout the experiments. First, they were provided with a 50 mmol l^{-1} sodium solution as their only water source for 1–8 weeks before the experiments and were given 120 mmol sodium intravenously 2 days before the experiment. Second, the high sodium concentration of the parotid saliva, associated with the tendency for the sodium concentration to plateau and to approach the maximum concentration (concentration at infinite flow) at relatively low flow rates, will only occur if the parotid glands of the kangaroos had a low reabsorptive capacity for sodium which, in turn, indicates that the animals were sodium-replete and had low mineralocorticoid levels. Third, the lack of change in the sodium/potassium ratio of the saliva when the kangaroos were pretreated with the aldosterone receptor blocker, spironolactone, also indicates that they were sodium-replete. Fourth, the high degree of linearity of the sodium secretion rate/flow regressions further indicates that there was no change in ductal sodium absorption and hence in mineralocorticoid levels throughout the experiments.

The potassium concentration of the kangaroo saliva was negatively correlated with flow, and this seems typical of saliva evoked by parasympathetic and parasympathomimetic stimulation. In some experiments, the potassium concentration rose again as the flow rate approached maximum. This phenomenon has been observed in other species (Petersen & Poulsen, 1967; Imai, Sueki & Yoshimura, 1970; Siegel, 1972) and is believed to result from the prolongation of salivary stimulation transients (Schneyer, Young & Schneyer, 1972).

Kangaroo parotid saliva had concentrations of chloride similar to those reported for sheep parotid saliva. The chloride concentration was always negatively correlated with flow rate, whereas the published relations between chloride concentration and flow for sheep parotid saliva are quite variable (Coats & Wright, 1957; Kay, 1960; Beal, 1979, 1980). There is a positive correlation between chloride and flow in saliva from the parotid glands of dog and man (Thaysen, Thorn & Schwartz, 1954; Brusilow & Cooke, 1959).

Positive correlations between salivary calcium concentration and flow, and negative correlations between salivary magnesium and flow, have been reported for salivas from other species (Young & Schneyer, 1981). However, in comparison to other species, the concentrations of calcium and magnesium in kangaroo parotid saliva were very low, with the calcium concentration being 25–30 % of that reported for sheep parotid saliva (Manas-Almendros, Ross & Care, 1982). Low concentrations of calcium and magnesium are to be expected, since high levels of phosphate in the presence of high pH would tend to cause salt precipitation in the salivary ducts if the levels of calcium and magnesium were not very low.

Kangaroo parotid saliva was always slightly hyposmotic with respect to plasma, although at high flow rates it was almost isosmotic. Increases in sodium, and particularly bicarbonate, concentrations were the major contributors to the increase in osmolal concentration with increasing flow rate. A positive correlation between saliva osmolality and flow has been reported for sheep parotid saliva (Beal, 1979) and for various hypotonic salivas (Burgen, 1955; Kostlin & Rauch, 1957; Brusilow & Cooke, 1959; Mangos & Braun, 1966).

The striking similarity in the inorganic composition of parotid saliva from sheep and red kangaroos is presumably an example of convergent evolution in the two separate groups of herbivores which these species represent and, as such, reinforces the belief that this type of saliva is a necessary adjunct to digestion by foregut fermentation.

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