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Sodium and potassium in blood and milk and plasma aldosterone levels in high-yield dairy cows

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Summary. Plasma sodium, potassium and aldosterone levels, daily milk production and milk sodium and potassium were measured in 10 Holstein × Friesian cows during a whole lactation period beginning in November and ending in November the following year. The milk production (4 p. 100 fat content) of these animals was 6 170 ± 66 kg (mean ± SEM). During the whole experimental period, the cows had free access to salt blocks and were thus always sodium-replete.

Plasma sodium and potassium levels showed no significant variations during lactation. The daily excretion of sodium and potassium through milk paralleled that of daily milk production. Plasma aldosterone levels decreased sharply from 77.4 ± 4.0 $pg.ml^{-1}$ at calving to 13.2 ± 3.6 $pg.ml^{-1}$ ($P < 0.01$) on day 7 of lactation, then remained stable until day 50 (16.4 ± 4 $pg.ml^{-1}$). They increased slightly on day 155 (60 days after mating : 36 ± 5 $pg.ml^{-1}$; $P < 0.05$) and abruptly after spring grazing (54.9 ± 11 $pg.ml^{-1}$; $P < 0.01$), then remained high until the end of lactation (48.5 ± 12 $pg.ml^{-1}$). Plasma aldosterone levels were 11.9 ± 2.4 $pg.ml^{-1}$ in seven 24-month old, non-pregnant heifers fed the same winter ration as the cows. No relationship could be demonstrated between sodium and potassium concentrations in blood and milk or between those parameters and plasma aldosterone levels.

Thus, in high-yield dairy cows, aldosterone does not seem to play a major role in the regulation of sodium and potassium excretion through milk.

Introduction.

Mean sodium and potassium concentrations in cows's milk are 22.5 $mmoles.l^{-1}$ and 38.5 $mmoles.l^{-1}$, respectively (Guéguen, 1971). Thus, for a cow producing 6 000 kg of milk per lactation, the simultaneous mammary output of sodium and potassium are 135 and 231 moles, respectively. The regulation of milk electrolyte concentration may be of importance in high-yield dairy cows.

The ionic composition of milk is more closely related to intracellular than to extracellular fluid. Mammary secretory cells have active, monovalent-cation pumps which increase potassium and decrease sodium only on the cell plasma membrane adjacent to the vascular supply (Johnson and Wooding, 1978). In contrast, the apical cell surface adjacent to the milk seems to be a simple, unselective permeability barrier to monovalent cations, thereby allowing the ratio of intracellular potassium/sodium to largely regulate that of milk (Falconer, 1980). Aldosterone is an important factor in sodium storage. However, results from studies on the possible influence of aldosterone on milk sodium concentration are conflicting. Using lactating guinea-pig mammary cells incubated *in vitro*, Linzell and Peaker (1971) could not demonstrate any effect of aldosterone on intracellular sodium and potassium concentrations, while Yagil, Etzion and Berlyne (1973) reported that the intraperitoneal injection of aldosterone (15 μg/kg body weight) in lactating rats caused a significant reduction in milk sodium concentration, and that that effect was inhibited by spironolactone. To our knowledge, nobody has studied the influence of aldosterone on the mineral composition of bovine milk. The purpose of the present work was to study the putative relationships between plasma aldosterone levels and sodium and potassium concentrations in the blood and milk of high-yield dairy cows.

Material and methods.

Animals. — Ten 6 to 8-year old Holstein × Friesian cows at their 4th to 6th calving were used during a lactation period. They were housed inside for 7 months after calving at the beginning of November, and fed maize silage and grain concentrate with vitamin and mineral complement so that, during the winter period, the daily ration of each contained approximately 25 g of sodium and 90 g of potassium. Since water and salt blocks (Oligobloc, Salins du Midi) were always available throughout the experi-

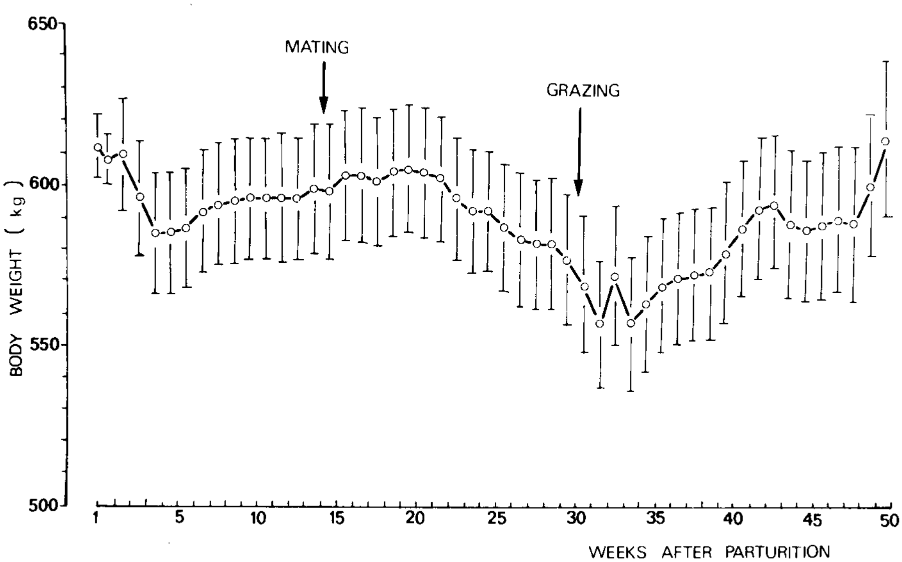

FIG. 1. — *Pattern of cow body weight during lactation* (mean ± SEM).

ment, the daily sodium and potassium intakes of each animal could not be measured, but all the cows were sodium-replete. They were moved to fescue pasture 7 months after calving and then ate grass until the drying-off period 49 weeks after calving. Each animal was weighed once a week during the experimental period (fig. 1), and they were artificially mated (after œstrus synchronization) 96 $\pm$ 18 days after calving.

The cows were mechanically milked twice a day at 6 a.m. and 4 p.m. throughout lactation, and the daily total milk production (2 milkings ; 4 p. 100 fat content) of each animal was measured (fig. 2). Milk samples were collected after the morning milking on each day of blood sampling and frozen until analysis.

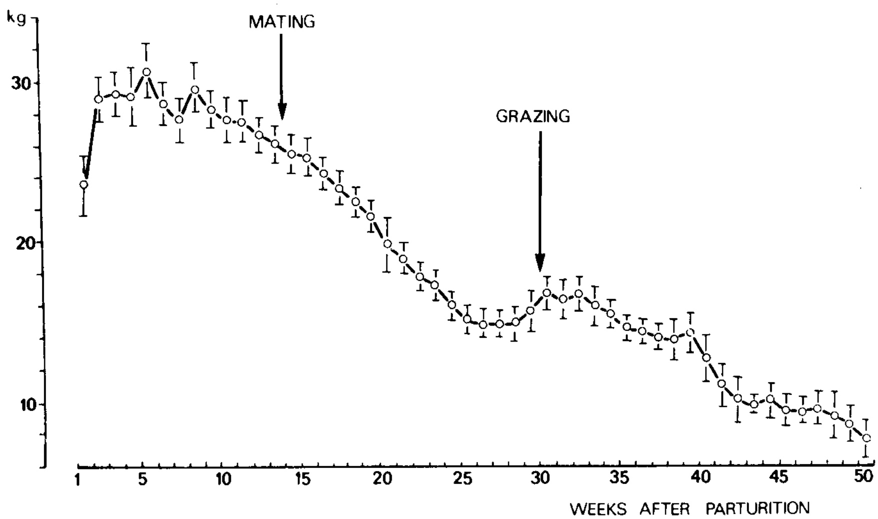

FIG. 2. — *Pattern of daily milk production* (4 p. 100 *fat content*) *during lactation* (mean $\pm$ SEM).

Serial blood samples (except those collected in the minutes following calving) were obtained at 8 a.m. by puncture of the jugular vein after milking. After measurement of the hematocrit (fig. 3) and centrifugation, the plasma was frozen until analysis.

Analyses. — Sodium and potassium concentrations were measured in thawed plasma samples and milk by flame emission (Perkin Elmer 420).

Plasma aldosterone levels were determined by radioimmunoassay (Bayard *et al.*, 1970 ; Giry and Delost, 1977). Thawed plasma samples were extracted with dichloromethane defatted at — 30 °C with 70 p. 100 methanol and centrifuged. Aldosterone was separated from cortisol and cortisone by paper chromatography (Bush B_5). Recovery, determined by radioactive (1,2-^{3}H)-aldosterone, was 80 p. 100. Method sensitivity was 20 pg and precision was 9 p. 100 for 100 to 400 pg.

Statistics. — Results are expressed as the mean $\pm$ SEM. Student's t-test was used for statistical comparison.

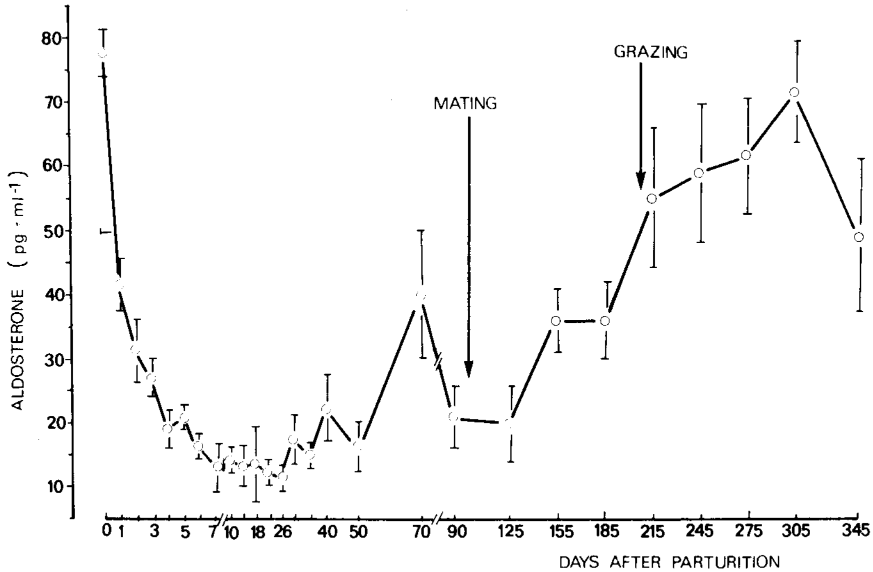

FIG. 3. — *Changes in hematocrit value during lactation* (mean ± SEM).

Results

The plasma sodium and potassium levels of the 10 cows showed no significant variation during the whole experimental period (fig. 4).

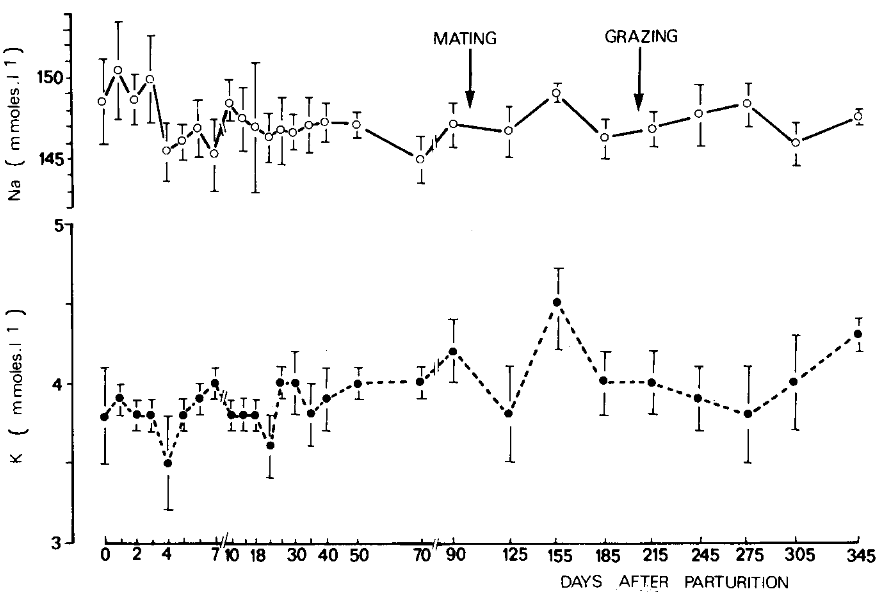

FIG. 4. — *Plasma sodium* (Na) *and potassium* (K) *levels in cows during lactation* (mean ± SEM).

Colostrum (or milk) sodium concentration decreased from 22.4 ± 1.1 mmoles.l^{-1} on day 1 after calving to 13.9 ± 1.4 mmoles.l^{-1} ($P < 0.01$) on day 30 after calving, then increased to 23.7 ± 3 mmoles.l^{-1} on day 50 and remained stable until the end of lactation. Milk potassium concentration remained stable from day 1 (42 ± 1.1 mmoles.l^{-1}) until day 70 (42.7 ± 1 mmoles.l^{-1}), then decreased until the end of lactation (34 ± 2 mmoles.l^{-1} ; $P < 0.05$) (fig. 5).

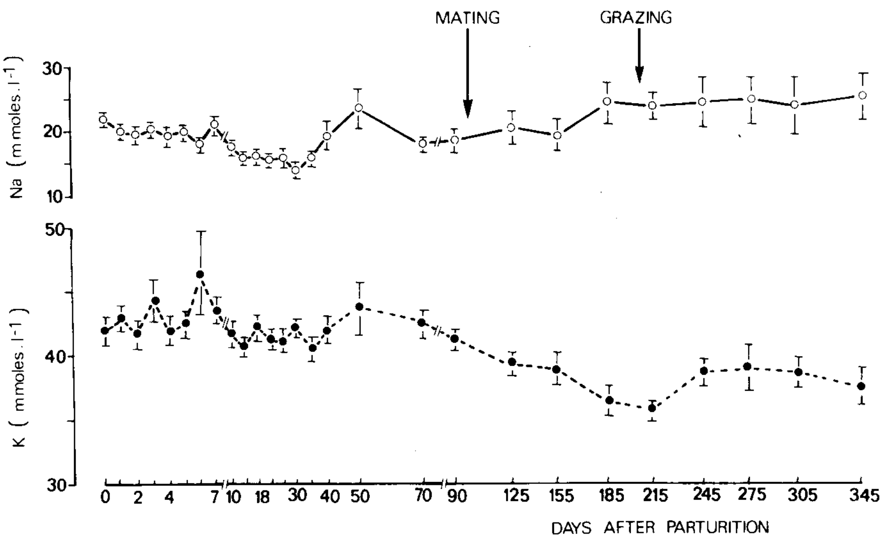

FIG. 5. — *Milk sodium* (Na) *and potassium* (K) *concentrations in cows during lactation* (mean ± SEM).

The daily excretion of sodium through milk decreased from 556 ± 35 mmoles on day 1 to 447 ± 33 mmoles ($P < 0.01$) during week 5, increased to 599 ± 35 mmoles ($P < 0.01$) during week 7, then decreased to 313 ± 41 mmoles ($P < 0.01$) during the last week of lactation. Daily excretion of potassium through milk increased from 942 ± 73 mmoles on day 1 to 1 300 ± 66 mmoles ($P < 0.01$) during week 7, then decreased to 471 ± 68 mmoles ($P < 0.01$) during the last week of lactation (fig. 6).

During the first week after parturition, plasma aldosterone levels decreased from 77.4 ± 4 pg.ml^{-1} immediately after calving to 13.2 ± 3.6 pg.ml^{-1} ($P < 0.01$) 168 hrs later, then did not significantly vary then until day 50 (16.4 ± 4 pg.ml^{-1}). They slightly increased on day 155 (60 days after artificial mating : 36 ± 5 pg.ml^{-1} ; $P < 0.05$) (fig. 7).

On the day before the cows were moved to spring pasture, plasma aldosterone levels were 34 ± 6 pg.ml^{-1}. They increased 24 hrs after grazing (53 ± 9 pg.ml^{-1}) ($P < 0.05$) and reached 71.5 ± 8 pg.ml^{-1} ($P < 0.01$) 58 days later, then remained high until the end of lactation (48.5 ± 12 pg.ml^{-1}) (fig. 7). In seven 24-month old, non-pregnant heifers, fed the same winter ration as the cows, plasma aldosterone levels were 11.9 ± 2.4 pg.ml^{-1}.

No relationship was found (i) between blood and milk sodium levels, (ii) between

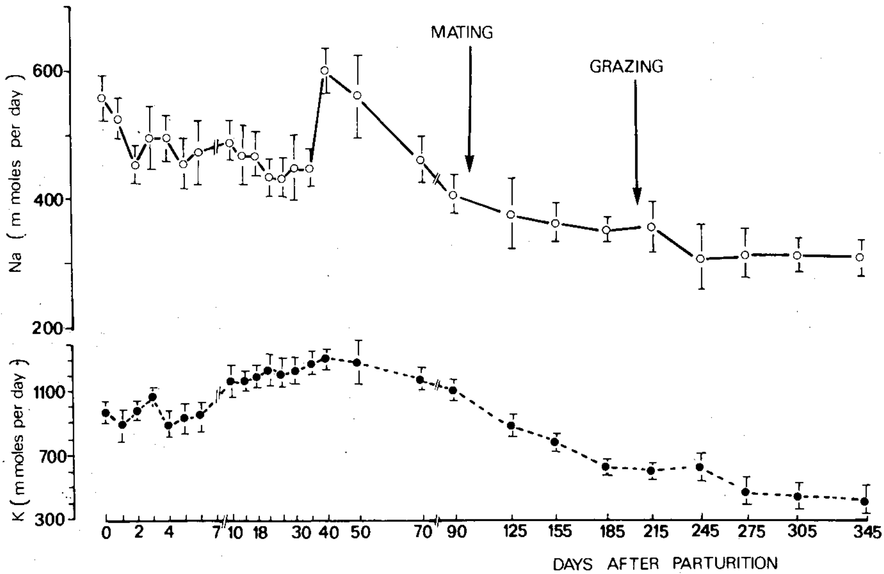

FIG. 6. — *Daily mammary excretion of sodium* (Na) *and potassium* (K) *during lactation* (mean ± SEM).

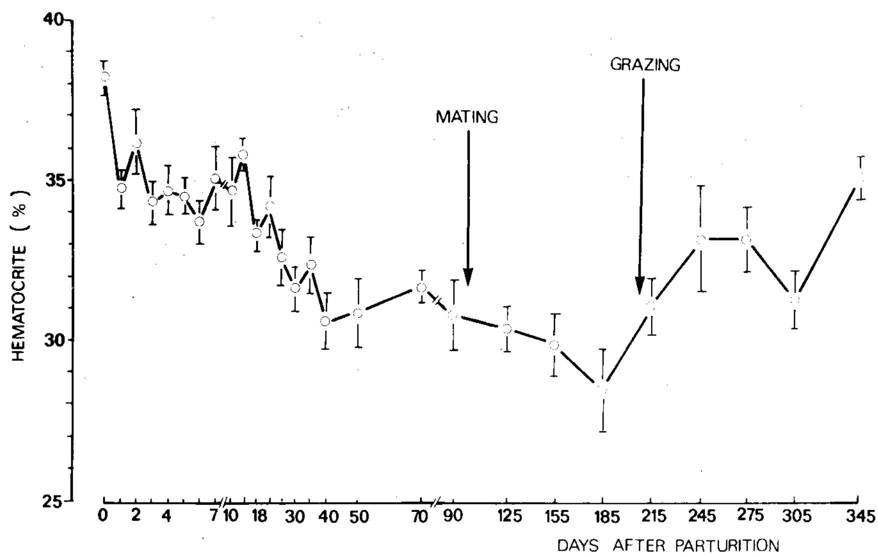

FIG. 7. — *Plasma aldosterone levels in cows during lactation* (mean ± SEM).

blood and milk potassium levels, (iii) between these electrolyte concentrations in blood or milk and plasma aldosterone levels, (iv) between plasma aldosterone levels and the hemocrit.

Discussion

The 10 animals used in this study produced 6 170 ± 66 kg of milk (4 p. 100 fat content) in one lactation lasting 346 ± 14 days. To our knowledge, such high-yield dairy cows have never been used to study the endocrine regulation of sodium and potassium metabolism.

During the first week after parturition, colostrum sodium concentration decreased, while that of potassium did not change (fig. 5) ; similar findings have been reported by Guéguen (1971). Our results concerning variations in milk sodium and potassium contents throughout lactation are similar to those reported by Guéguen and Journet (1961) in 10 cows of different breeds between weeks 3 and 40 of lactation. In the course of lactation, milk potassium content diminished regularly, whereas its sodium content increased (fig. 5). However, daily milk production seems more effective than milk mineral content in determining the daily sodium and potassium excretion through the mammary gland, which parallels that of daily milk production (figs. 2, 6). As reported by Guéguen and Journet (1961), moving the cows from winter feeding to spring pasture did not change milk mineral composition.

Plasma sodium and potassium levels remained constant during the whole lactation period (fig. 4). No relationship between plasma sodium and aldosterone levels was evident in our animals. Plasma sodium and potassium concentrations were similar to those already reported in lactating and pregnant cows (Lumsden, Mullen and Rowe, 1980 ; Wilson *et al.*, 1980). In the Holstein × Friesian cows in the present study, plasma aldosterone levels varied between 20 and 100 pg . ml^{-1} (fig. 7). The mean plasma aldosterone level in 144 Holstein cows, measured by double-antibody radioimmunoassay, was 67.4 ± 41.5 pg . ml^{-1} (Bardwell *et al.*, 1978). In 11 young Holstein × Friesian calves, plasma aldosterone levels varied between 119 ± 7 pg . ml^{-1} at birth and 13 ± 5 pg . ml^{-1} at 7 days (Safwate *et al.*, 1981). Plasma aldosterone levels measured in adult guinea-pigs (Giry and Delost, 1977), mice (Dalle *et al.*, 1978) and mares (Giry *et al.*, 1979) were about 400 pg . ml^{-1}, 350 pg . ml^{-1} and 100 pg . ml^{-1}, respectively. Thus, the bovine species seems to be characterized by low plasma aldosterone levels.

In our experimental conditions, it was not possible to separate the possible influence of lactation from that of pregnancy since the cows were artificially mated around week 13 of lactation. Plasma aldosterone levels decreased sharply during the week after parturition (fig. 7). Similarly, plasma aldosterone levels decreased after delivery in ewes (Boulfekhar and Brudieux, 1980 ; Moncaup *et al.*, 1980), mares (Giry *et al.*, 1979), guinea-pigs (Giry and Delost, 1977), mice (Dalle *et al.*, 1978) and women (Lammintausta and Erkkola, 1977). Although the metabolic clearance rate of aldosterone in ewes increased after parturition, the changes observed in the plasma levels of this hormone during the perinatal period in those animals could not be directly related to modifications in its catabolism (Moncaup *et al.*, 1980). According to Boulfekhar and Brudieux (1980), the increase in plasma aldosterone observed at the end of pregnancy is largely due to the stimulation of renin substrate production by oestrogens whose values are high during that period (Thorburn *et al.*, 1972). Thus, it is conceivable that the abrupt decrease in plasma oestrogen levels, occurring immediately after parturition in cows (Comline *et al.*, 1974), could induce a decrease in aldosterone secretion.

High aldosterone levels during pregnancy might prevent excessive salt loss. However, in pregnant women, plasma aldosterone concentration is positively correlated with plasma sodium level, indicating that sodium is probably not of major importance in the control of aldosterone secretion in pregnancy (Weir *et al.*, 1975). Similarly, normal aldosterone secretion rate has been measured in pregnant bitches which escape the sodium retention induced by deoxycorticosterone acetate, suggesting the existence of sodium-retaining mechanisms other than that provided by high plasma aldosterone levels (Robb *et al.*, 1970). Similar conclusions have been reached recently in pregnant rats (Churchill, Bengele and Alexander, 1980). In lactating pregnant cows, no relationship could be demonstrated between plasma aldosterone levels (fig. 7) and sodium and potassium concentrations in milk (fig. 5) or the excretion of these electrolytes through milk (fig. 6). Thus, in salt-replete cows, aldosterone does not seem to play a major role in the regulation of mammary sodium and potassium excretion. Milk sodium and potassium concentrations are probably mainly regulated by prolactin (Falconer, 1980). In mammary tissue incubated *in vitro*, prolactin prevents intracellular ion leakage by tightening the cell junctions (Linzell, Peaker and Taylor, 1975). Prolactin also activates the reciprocal transport of Na/K in mammary tissue *in vivo* (Falconer and Rowe, 1977).

When our cows ate young graminaceous grass (*Festuca pratensis*), their plasma aldosterone levels rose (fig. 7). This grass is very poor in sodium (0.25 g per kg dry matter) and rich in potassium (30 g per kg dry matter) ; its potassium content decreases regularly from the first vegetative cycle in May to the fourth one in October (Guéguen and Fauconneau, 1961). The circulating renin increases in sheep with a progressive sodium defect, leading to an increase in angiotensin II proportional to aldosterone secretion and to peripheral aldosterone concentration (Blair-West *et al.*, 1970). In pregnant ewes in which KCl infusion increased plasma potassium of 0.4-1.0 mmoles.l^{-1}, aldosterone increment was 10 ± 5 ng.dl^{-1} (Wintour *et al.*, 1979). In 18-month old dairy heifers, the ingestion of a potassium-rich diet (30 g K per kg of fresh matter) induced a significant rise in plasma aldosterone levels (Safwate and Barlet, 1981). Thus, the rise in plasma aldosterone levels after grazing on spring pasture probably resulted from both an increased intake of potassium and a decreased intake of sodium.

No relationship could be established between the fluctuations in the hemocrit values (fig. 3) and those of aldosterone (fig. 7).

There was no evident relationship between milk sodium or potassium concentration and plasma aldosterone levels in high-yield, sodium-replete dairy cows, indicating that aldosterone did not play a major role in regulating the excretion of these electrolytes through milk. However, grazing lush spring grass was associated with a significant increase in plasma aldosterone levels, possibly mediated by an increase in dietary potassium.

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Résumé. La concentration du sang et du lait en sodium et en potassium, et l'aldostéronémie, ont été mesurées durant une lactation chez 10 vaches Holstein × Frisonnes, dont chacune a produit au cours de celle-ci 6 170 $\pm$ 66 kg de lait à 4 p. 100 de matière grasse. Au cours de la période expérimentale (commencée début novembre et terminée fin novembre suivant) les animaux ont toujours eu libre accès à des pierres à sel.

La natrémie et la kaliémie des animaux ne varient pas pendant la lactation et la gestation. L'excrétion journalière de sodium et de potassium dans le lait suit les évolutions de la production laitière journalière. L'aldostéronémie est caractérisée par une décroissance très rapide entre le moment de la parturition (77,4 $\pm$ 4 pg.ml^{-1}) et le 7^{e} jour de lactation (13,2 $\pm$ 3,6 pg.ml^{-1} ; $P < 0{,}01$). Elle reste ensuite stable jusqu'au 50^{e} jour (16,4 $\pm$ 4 pg.ml^{-1}) augmente légèrement au 155^{e} jour (60 jours après insémination : 36 $\pm$ 5 pg.ml^{-1} ; $P < 0{,}05$), et fortement au moment de la mise à l'herbe des animaux (54,9 $\pm$ 11 pg.ml^{-1} ; $P < 0{,}01$) pour rester élevée jusqu'à la fin de la lactation (48,5 $\pm$ 12 pg.ml^{-1}). Chez 7 génisses témoins de 24 mois non gestantes, recevant la même ration alimentaire hivernale que les vaches, l'aldostéronémie est de 11,9 $\pm$ 2,4 pg.ml^{-1}.

Aucune corrélation n'a pu être mise en évidence entre les concentrations de sodium et de potassium dans le sang et dans le lait, ni entre ces paramètres et l'aldostéronémie. Ainsi, chez la Vache, l'aldostérone ne semble pas jouer un rôle majeur dans la régulation de l'excrétion mammaire du sodium et du potassium.

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