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FLUCTUATIONS IN THE GLUCOSE LEVEL OF COW'S MILK FROM NORMAL AND SUBCLINICALLY DISEASED UDDERS

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ABSTRACT

GIESECKE, W. H., DURAND, ANETTE M. & PETZER, INGE-MARIÉ, 1984 Fluctuations in the glucose level of cow's milk from normal and subclinically diseased udders. *Onderstepoort Journal of Veterinary Research*, 51, 15–19 (1984)

Individual quarter samples from some 19 cows on average were investigated monthly over 12 months for determining the udder health status of cows and the glucose concentrations of foremilk and strippings.

Foremilk showed a mean 0,1311 mM concentration of glucose which remained fairly stable during the period of investigation and lactation. A fluctuating mean value of 0.2037 mM was determined in strippings in which glucose levels were consistently and appreciably higher than those of foremilk.

Foremilk from completely normal quarters and others affected by non-specific cellular reaction, relevant or irrelevant teat canal infection and aseptic or septic subclinical mastitis, showed mean glucose concentrations of 0,1410; 0,1392; 0,1337; 0,1417; 0,1262 and 0,1248 mM, respectively. Strippings from the same quarters showed corresponding values of 0,2056; 0,2861; 0,2100; 0,1733; 0.1661 and 0,1617 mM glucose.

INTRODUCTION

Glucose is central to the functioning of the bovine mammary gland (Bauman & Davis, 1974; Davis & Bauman, 1974; Ebner & Schanbacher, 1974; Saacke & Heald, 1974). From the very limited available data, lacteal levels of glucose may apparently range from some 0,07–1,03 mM, depending on the methods of determination used and the lactational health status of the udders investigated (Reineccius, Kavanagh & Keeney, 1970; Mackie, Giesecke, Lück & De Villiers, 1977; Faulkner, Chaiyabutr, Peaker, Carrick & Kuhn, 1981; Giesecke & Van den Heever, 1981; Lück & Botha, 1982). Though levels of glucose in secretions from normal and abnormal udders may seem rather low, they could nevertheless be of significance to the leucocytic udder barrier.

The data available clearly suggest that glucose is essential for milk secretion and especially for the secretion of lactose and the aqueous phase of milk (Linzell & Peaker, 1971; Ahrné, Björck & Claesson, 1983). Concentrations of glucose in milk apparently reflect those within the mammary cell, since the former equate with the latter (Faulkner *et al.*, 1981) across the apical surface of the secretory epithelium. By contrast, the basal surface of the secretory epithelium seems subject to factors that, in spite of high levels of glucose in blood (i.e. ± 3,26 mM), limit the intra-epithelial concentration of glucose to some 0,2 mM.

From other data it is apparent that glucose also is essential for the normal functioning of polymorphonuclear neturophilic (PMN) leucocytes. They depend for the killing of phagocytized bacteria on several bactericidal mechanisms, including the myeloperoxidase-halide-hydrogen peroxide system, which facilitates the iodination reaction (Karnovsky, 1962; Stossel, 1974; Klebanoff, 1975). PMN-leucocytes in milk are specially important for protecting the bovine udder from infection. Significant correlations of PMN-leucocytes and glucose in different udder secretions have been reported (Mackie *et al.*, 1977; Giesecke & Van den Heever, 1981).

Availability, uptake and use of glucose on the level of the mammary epithelium and the PMN-leucocytes in milk may change during stress which therefore seems a particularly important factor in bovine udder health.

Stress conditions, such as milk fever of dairy cows (Hayashi, Ono, Sato & Miyake, 1979), are associated with increased plasma levels of glucocorticoids. The latter *inter alia* are known to affect cellular uptake and intracellular metabolism of glucose. Mammary epithelium apparently has a high affinity for glucocorticoid (Gorewit & Tucker, 1977; Collier & Tucker, 1978; Pope & Swinburne, 1980). It further limits the concentration in milk of glucocorticoid to levels which, though lower than those in plasma (Schwalm & Tucker, 1978; Hayashi *et al.*, 1979; Pope & Swinburne, 1980; Fox, Butler, Everett & Natzke, 1981), nevertheless are positively correlated with the latter (Gwazdauskas, Paape & McGilliard, 1977; Bremel & Gangwer, 1978; Fox *et al.*, 1981).

Elevated concentrations of glucocorticoid in both normal and mastitic mammary secretions have been observed after parenteral administration of synthetic glucocorticoid and ACTH (Gwazdauskas *et al.*, 1977; Fox, Heald, Gwazdauskas & Vinson, 1981; Paape, Gwazdauskas, Guidry & Weinland, 1981; Tainturier, Alvinerie, Brandon & Toutain, 1982). They indicate that, during stress, susceptibility to mastitis may increase owing to the directly or indirectly impaired functioning of lymphocytes and PMN-leucocytes (Paape *et al.*, 1981; Roth, Kaeberle & Hsu, 1982). The latter workers showed that a decreased iodination reaction is related to a significantly affected myeloperoxidase-halide-hydrogen peroxide system.

It is clear from the above investigations that interactions of stress, lacteal levels of glucocorticoid and glucose and their effect on the functioning of the leucocytic udder barrier can be of great significance to bovine udder health. They probably also affect the development of acute clinical mastitis during cold weather (Schildbach, 1960; Hropot, 1970) and may be associated with further problems of milk production of dairy cattle subject to diverse stressors (Bianca, 1965; Lee, Beatty & Roussel, 1971; Smith, Edgerton, Hafs & Convey, 1973; Thompson, 1973; Guidry, Paape & Pearson, 1976; Paape, Desjardins, Guidry, Miller & Smith, 1977; Hayashi *et al.*, 1979; Ingraham, Stanley & Wagner, 1979; Gwazdauskas, Paape, Peery & McGilliard, 1980).

In the light of the foregoing conclusions further work on glucose in milk from cows with different states of udder health seems warranted.

MATERIALS AND METHODS

Experimental animals

The investigation was conducted on a total of 30 grade Friesian cows that differed in age, number of lactations, daily milk yield and stage of lactation. The cows were generally healthy, in good condition and free from brucellosis and tuberculosis. They were fed, kept, handled and milked under routine conditions. Average production per cow was approximately 3 400 kg per lactation and 10 kg of milk per day.

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Depending on their stage of pregnancy during the 12 months of the investigation, the cows commenced, advanced and eventually completed their lactations on different dates. Of the 30 cows involved, an average of 19 cows were tested during any of the investigations (Table 1), whereas the individual cow on average was subjected to 8 successive investigations performed at monthly intervals.

The cows were milked by machine at 08h00 and 14h00 every day. The milking routine included washing of teats with lukewarm disinfectant solution and drying them thoroughly with individual disposable paper towels before attachment to the milking machine. Immediately after milking the teats were submerged in disinfectant teat dip solution.

Collection and examination of milk samples

The investigation was performed on some 950 samples each of foremilk and strippings collected once a month.

Samples of foremilk and strippings were aseptically collected from individual quarters during the morning milking. The samples were processed for the determination of bacterial growth, electronic cell counting, bovine serum albumin and glucose as described by Giesecke & Van den Heever (1981), except that 0,6 N perchloric acid was used instead of uranyl acetate for the deproteinization of samples.

Evaluation of results

The state of udder health of quarters was evaluated on foremilk on the clinical appearance of their secretion and the laboratory criteria proposed by Giesecke & Viljoen (1974). Because the same cows and quarters were repeatedly examined no investigation was performed on a random sample of cows/quarters. Statistical analyses were thus limited to calculations of basic statistical values (e.g. arithmetic mean, standard error of mean, etc.) required for assessing general trends.

RESULTS

Levels of glucose at different periods of investigation and lactation

During the investigation, concentrations of glucose (mM) showed mean values ($\bar{x}$), and standard errors of the mean ($SE\bar{x}$) which, for foremilk and strippings, amounted respectively to 0,1311 ($\bar{x}$), and 0,0015 ($SE\bar{x}$) and 0,2037 ($\bar{x}$), and 0,0051 ($SE\bar{x}$).

Irrespective of the period of investigation (Fig. 1) and lactation (Fig. 2), glucose concentrations in foremilk seemed fairly stable, whereas those of strippings tended to fluctuate, especially during the initial months of investigation (Fig. 1) and more advanced periods of lactation (Fig. 2).

The data (Fig. 1 & 2) further indicate that strippings showed mean concentrations of glucose consistently and appreciably higher than those of foremilk. However, all such data were apparently subject to considerable cow to cow variations as indicated by the corresponding SD and $SE\bar{x}$ values (Tables 2 & 3).

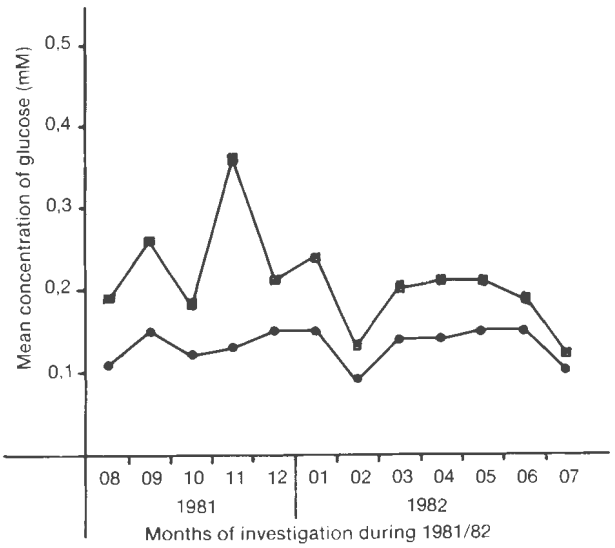

FIG. 1. Levels of glucose in foremilk (•——•) and strippings (■——■) at different periods of investigation

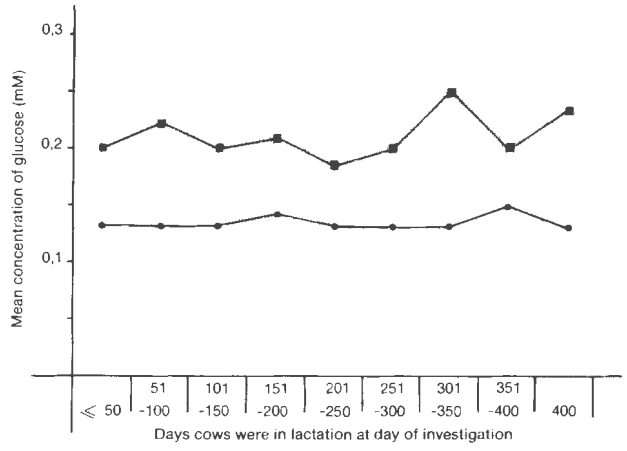

FIG. 2. Levels of glucose in foremilk (•——•) and strippings (■——■) at various periods of lactation

TABLE 1 The numbers of cows per class of days in lactation at the time of their investigation

Days in lactation	Year/month of investigation/corresponding numbers of cows investigated											
	1981					1982						
	08	09	10	11	12	01	02	03	04	05	06	07
>400	5	3	4	3	1	1	1	1				
351–400	3	1				1				1	1	
301–350			2	4	2							2
251–300	4	5	3						1	4	7	4
201–250	7				1	2	1	2	8	3	5	3
151–200				2	1	2	3	7	4	6	3	3
101–150	1	1	2	2	2	7	6	6	3	3	2	1
51–100	3	2	2	3	6	5	5	3	3	1		1
=<50	4	3	6	4	6	3	2	1		1	1	2
Total.....	27	15	19	18	19	21	18	20	19	19	19	16

TABLE 2 Glucose values ($\bar{x}$, SD, SE$\bar{x}$) of foremilk and strippings analysed during various months of investigation

Month/year of investigation	Foremilk Glucose values (mM)			Strippings Glucose values (mM)		
	$\bar{x}$	SD	SE$\bar{x}$	$\bar{x}$	SD	SE$\bar{x}$
08/81	0,1143	0,0619	0,0048	0,1881	0,1646	0,0130
09/81	0,1527	0,0540	0,0069	0,2551	0,1602	0,0209
10/81	0,1211	0,0245	0,0028	0,1764	0,1202	0,0141
11/81	0,1313	0,0241	0,0028	0,3615	0,3169	0,0396
12/81	0,1493	0,0700	0,0080	0,2066	0,0903	0,0106
01/82	0,1536	0,0287	0,0031	0,2384	0,0994	0,0112
02/82	0,0935	0,0169	0,0020	0,1310	0,0377	0,0045
03/82	0,1376	0,0435	0,0049	0,2017	0,1149	0,0145
04/82	0,1422	0,0336	0,0039	0,2051	0,1042	0,0139
05/82	0,1462	0,0434	0,0050	0,2056	0,1096	0,0153
06/82	0,1450	0,0301	0,0035	0,1943	0,1006	0,0115
07/82	0,0987	0,0132	0,0017	0,1088	0,0272	0,0035

TABLE 3 Glucose values ($\bar{x}$, SD, SE$\bar{x}$) of foremilk and strippings analysed during different periods of lactation

Periods of lactation (days)	Foremilk Glucose values (mM)			Strippings Glucose values (mM)		
	$\bar{x}$	SD	SE$\bar{x}$	$\bar{x}$	SD	SE$\bar{x}$
=<50	0,1298	0,0585	0,0049	0,2001	0,1523	0,0131
51–100	0,1282	0,0355	0,0030	0,2184	0,1736	0,0147
101–150	0,1314	0,0386	0,0032	0,1988	0,1198	0,0105
151–200	0,1388	0,0426	0,0039	0,2068	0,1143	0,0114
201–250	0,1253	0,0410	0,0033	0,1749	0,0849	0,0074
251–300	0,1297	0,0524	0,0046	0,1958	0,1429	0,0133
301–350	0,1328	0,0568	0,0090	0,2497	0,2981	0,0471
351–400	0,1515	0,0587	0,0104	0,2030	0,1234	0,0242
>400	0,1300	0,0534	0,0062	0,2279	0,1934	0,0236

TABLE 4 Values of glucose ($\bar{x}$, SD, SE$\bar{x}$) in foremilk from udder quarters with different subclinically determinable udder health states

Udder health states determined	Number of samples*	Glucose values (mM)		
		$\bar{x}$	SD	SE$\bar{x}$
Completely normal	73	0,1410	0,0686	0,0080
Non-specific cellular reaction	6	0,1392	0,0330	0,0135
Irrelevant teat canal infection	341	0,1337	0,0488	0,0027
Relevant teat canal infection	41	0,1417	0,0618	0,0097
Aseptic mastitis	17	0,1262	0,0267	0,0065
Septic mastitis	101	0,1248	0,0357	0,0036

* The difference between samples indicated and investigated as a whole is due to the exclusion of samples/quarters not clearly conforming with the diagnostic criteria used for determining the states of udder health

TABLE 5 Values of glucose ($\bar{x}$, SD, SE$\bar{x}$) in strippings from udder quarters in various subclinical health states diagnosed in terms of investigations performed on corresponding foremilk samples*

Udder health states determined	Number of samples	Glucose values (mM)		
		$\bar{x}$	SD	SE$\bar{x}$
Completely normal	73	0,2056	0,0967	0,0115
Non-specific cellular reaction	6	0,2861	0,2044	0,0836
Irrelevant teat canal infection	341	0,2100	0,1650	0,0095
Relevant teat canal infection	41	0,1733	0,0939	0,0154
Aseptic mastitis	17	0,1661	0,0989	0,0248
Septic mastitis	101	0,1617	0,0861	0,0091

* Samples referred to in Table 4

From the above data it seems conceivable that glucose concentrations, determined at intervals shorter than those used at present, may show greater fluctuations from investigation to investigation than those indicated in Fig. 1 & 2.

Levels of glucose in foremilk and strippings from quarters with different udder health states determinable subclinically

Glucose concentrations of samples of foremilk and strippings differed according to the health status of their quarters of origin (Tables 4 & 5).

In foremilk (Table 4), the highest glucose concentrations with remarkably similar values were found in milk from quarters either completely normal or those affected by relevant teat canal infection. Intermediate levels of glucose, some 3,33 % lower than the former, were determined in milk from quarters with non-specific cellular reaction and irrelevant teat canal infection. The lowest and least variable glucose concentrations, differing from the highest and intermediate ones by some 11,00 % and 7,77 % respectively, were noted in samples of mastitic quarters.

It is noteworthy that milk from subclinical mastitic quarters showed decreased glucose concentrations and thus indicated a tendency similar to but more limited than that already described for acute clinical mastitis (Giesecke & Van den Heever, 1981). It is equally interesting that relevant teat canal infection and subclinical septic mastitis proper, diagnosed as 1 condition (i.e. sublcinical mastitis) by means of generally acknowledged diagnostic criteria (Tolle, 1971), were related to different mean levels of glucose.

In the strippings (Table 5) of the same quarters the udder health states were associated with glucose concentrations markedly higher and more variable than the corresponding values in foremilk. Compared with the latter, strippings of completely normal quarters and others affected by non-specific cellular reaction, irrelevant or relevant teat canal infection, aseptic or septic mastitis, showed glucose concentrations elevated by 45,82; 105,53; 57,07; 17,18; 31,62 and 29,57 % respectively. Correspondingly, the glucose values of strippings differed by +0,93; +40,45; +3,09; −14,98; −18,46 and −20,62 % from the mean value of 0,2037 mM glucose determined for all strippings.

DISCUSSION

The glucose values of the foremilk (i.e. $\bar{x}$ = 0,1311 mM) and strippings (i.e. $\bar{x}$ = 0,2037 mM) investigated were within the range of glucose concentrations found in milk from goat, cow, sheep and rabbit (Faulkner *et al.*, 1981). Those of foremilk alone were similar to levels also determined by Giesecke & Van den Heever (1981) and Ahrné *et al.* (1983), but appreciably below the range of mean values of 0,77–1,03 mM reported by Reineccius *et al.* (1977) and Lück & Botha (1982). Such variations may be due to the different methods used for determining lacteal levels of glucose.

The data (Fig. 1 & 2) further indicate that, during the months of investigation and lactation, glucose concentrations in foremilk remained fairly stable, whereas those of strippings tended to fluctuate. The limited change in the glucose concentrations in foremilk was rather deceptive. Shorter investigation intervals may indicate greater fluctuations which possibly depend on factors, such as abnormal udder health (Giesecke & Van den Heever, 1981) and sudden adverse weather conditions (Giesecke & Arnold, unpublished data, 1981) affecting dairy cows. It also remains uncertain whether the fluctuating glucose values of strippings (Fig. 1) were actually due to seasonal factors or to more lactational ones (Fig. 2), because at the time such fluctuations were most distinct an appreciable number of the cows investigated were either at the beginning or at an abnormally advanced stage of lactation (Table 1).

More detailed data (Tables 2 & 3) on the former (Fig. 1) and latter results (Fig. 2) confirm the finding of Lück & Botha (1982) that considerable cow to cow variations of lacteal glucose concentrations occur, but they do not indicate a slight though significant increase of lacteal glucose levels during lactation observed by these workers. However, the present investigation suggests that glucose concentrations of foremilk and strippings may differ more often than observed by Lück & Botha (1982), and depend, for example, on factors such as level of complet milking and massaging of udders at the sampling of strippings.

Data on the secretion of lactose and milk fluid (Linzell & Peaker, 1971) suggest the intramammary equilibration of glucose (Faulkner *et al.*, 1981), and the increased secretional activity and deterioration of mammary epithelium at the end and during the early milking intervals (Linzell, 1960, 1974; Hollman, 1974; Peaker, 1978; Saacke & Heald, 1974) seem to indicate that consistent elevated glucose levels are possible in strippings.

Conversely, glucose levels in strippings may be especially subject to conditions, such as stress, starvation (Linzell & Peaker, 1971; Faulkner *et al.*, 1981), changes of glucocorticoid in plasma and milk and other factors causing mammary regression (Lascelles & Lee, 1974), which decrease the secretional activity of mammary epithelium. Mammary regression, induced artificially, resulted in significantly reduced lacteal concentrations of glucose (Mackie *et al.*, 1977). Such a decrease of glucose values seems especially interesting in relation to mastitis and the functioning of the leucocytic udder barrier.

Several histophathological investigations suggest that, during mastitis, inflammatory changes are almost automatically associated with changes similar to those of accelerated, induced regression (Chandler, 1970; Reid, Harrison & Anderson, 1976; Heald, 1979; Nickerson & Heald, 1982). Mackie *et al.*, (1977) showed that induced regression may be related to reduced glucose concentrations in milk. Decreased glucose levels in acute clinical mastitis, discussed by Giesecke & Van den Heever, (1981), may thus be related not only to inflammatory but also to simultaneous regressive intramammary changes. It seems conceivable, therefore, that the lower concentrations of glucose observed during the present investigation in milk from subclinically mastitic quarters (Tables 4 & 5) also may result from inflammatory and regressive tissue changes.

The reduced mean concentration of glucose in subclinical mastitis and the values related to teat canal infection (Tables 4 & 5) further indicate that the 2 udder health states may not only differ in their diagnostic criteria (Giesecke & Viljoen, 1974), but also in factors determining their respective concentrations of glucose in foremilk and strippings.

The present investigation was mainly undertaken to arrive at a more complete general understanding on the significance for bovine udder health of glucose in milk. Previous results (Mackie *et al.*, 1977; Giesecke & Van den Heever, 1981) and the present ones do not in any way support the suggestion of Lück & Botha (1982) that "cows producing milk with a high glucose content should be more resistant against mastitis than cows producing milk with a low glucose content". Further work on the significance of lacteal glucose in bovine udder health seems essential.

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