Detection of Abnormal Quarter Milk by the Quarter Difference of the Electrical Conductivity and Its Theoretical Basis

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It appears reasonable to assume that the chemical composition of milk taken from each quarter of individual udder of a cow is same from the physiological point of view. This, however, is not necessarily true when it is examined. Except heifers in their first lactation, some diversity is commonly found among quarter milks taken from individual cows. The author has examined the diversity for years through a routine sampling of quarter milks of different cows, using a separate quarter milking machine, and analysing chemical composition of them. Among the milk constituents the concentrations of sodium, potassium and chloride as the major electrolytic components, and the content of fat, protein and lactose were measured routinely, and the contents of the electrolytes and of lactose were found most useful for the purpose.

In the course of the study, using cows from the Institute herd and in addition some cows from dairy farms, it was found that about 1/3 of cows showed abnormality in some of their quarter milks, and that the abnormal milk appeared only in one or two quarters in about 90% of these cows, and further that the abnormal milk appeared repeatedly in the same quarter or quarters of the cow[1].

The abnormal quarter milk observed in the study showed higher sodium and chloride and lower lactose and potassium concentration than the supposedly normal quarter milk in the same udder. In addition, the milk yield of the quarter that the former milk was taken was less than the contra-lateral normal quarter

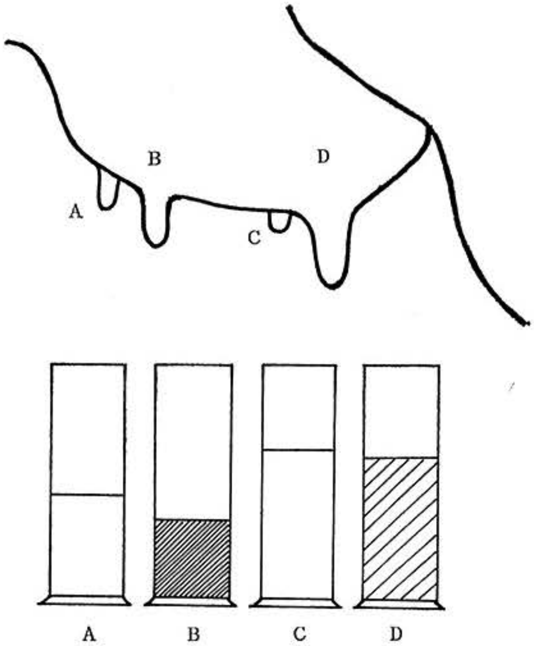

Fig. 1. Diversity of milk composition in quarter milks of individual udder, that is commonly found in milking cows; the cause is attributable broadly to subclinical mastitis. The milk composition of the quarter B milk is abnormal and D is slightly abnormal. The milk yields of these quarters are less than the contra-lateral quarters.

(Fig. 1).

Such changes in milk composition and in yield had been known as occur in the quarter suffered from mastitis. Therefore, it seems probable that the anomalousness of the quarter which produces abnormal milk bears a close relation with what is called subclinical mastitis.

Indirect methods for detecting abnormal milk without bacterial examinations

In 1941, Vanlandingham et al.[2] reported a method for detecting subclinical mastitic milk. The method was based on comparisons of the quarter milks taken from individual udders. It was one of what is called indirect methods which dispense with bacterial examinations of milk. They chose a normal quarter milk out of the four of an udder, by comparing concentrations of a constituent, for instance chloride, and calculated the difference between each of quarter milk and the normal one. This is called the quarter difference value. In order to sort out the abnormal quarter milk from the rest, they settled a criterion by using a statistical method. They calculated the mean and the standard deviation from a large numbers of quarter difference values that were obtained from several healthy milking cows (6 quarter difference values an udder), and made the value, (means)+2×(s.d.), the criterion for judgement.

According to the principle mentioned above, the author reexamined which was the most suitable criterion for detecting abnormality of quarter milk. Efficacies of the quarter differences in sodium, chloride, potassium, lactose and in addition, some combinations of these components were compared for the purpose, and it was found the quarter difference of sodium plus chloride, (Na+Cl) value, was most effective. The criterion for the abnormality judgement by the quarter difference of Na+Cl value, hereinafter referred to Qd(Na+Cl), was 7.2 mEq/l. The value was calculated from 368 quarter milk samples taken from 20 cows from the Institute herd. When the difference in (Na+Cl) value between the quarter milk with the lowest Na+Cl value and any other quarter milk or milks of an udder exceeds 7.2 mEq/l, the latter should be graded abnormal[1].

Abnormal quarter milk and its electrical conductivity

Sodium, chloride and lactose are the solutes, being different from fat or protein which are suspended in milk, and osmolarity of milk is largely determined by the former three substances and potassium. The milk osmolarity is in a continuous equilibrium with the blood plasma osmolarity, and which is kept virtually constant by the homeostatic mechanism of living animals. Therefore, the milk osmolarity is unchangeable, whether it is normal or abnormal milk as the mastitic milk. If an increase of Na, which is always associated with an increase of Cl, occurs in an abnormal milk, the decreases of other milk constituent counterbalance it in order to keep the constancy of milk osmolarity.

Interrelationships between the increases and the decreases in various milk constituents in the abnormal quarter milk were therefore studied, and simple correlations expressible by linear regression equations were found[3]. The relationship between the increase of Na, that is, the quarter difference of Na, and concomitant increase of Cl in the abnormal milk was rectilinear, and the ratio of the two was similar to the ratio of the concentration of the two in the blood plasma.

The relationships between the increase of Na+Cl and the decrease of K, lactose (L) and K+L were rectilinear as shown in Fig. 2. The figure also indicated that the decrease in K+L (m mol/l) corresponded well over 80% with the increase of Na+Cl (mEq/l), and therefore, it appeared that the rise in osmolarity in the abnormal milk due to the increases of Na and Cl was counterbalanced mainly by the decreases of lactose and K.

The contribution of the major solutes to the osmolarity of normal and abnormal milk is shown diagrammatically in Fig. 3. In abnormal milk, lactose, a non-electrolytic solute, decreases, chiefly against the increase of Na^+ and Cl^-, with a relatively small decrease, about 1/4 of Qd(Na+Cl), of K^+.

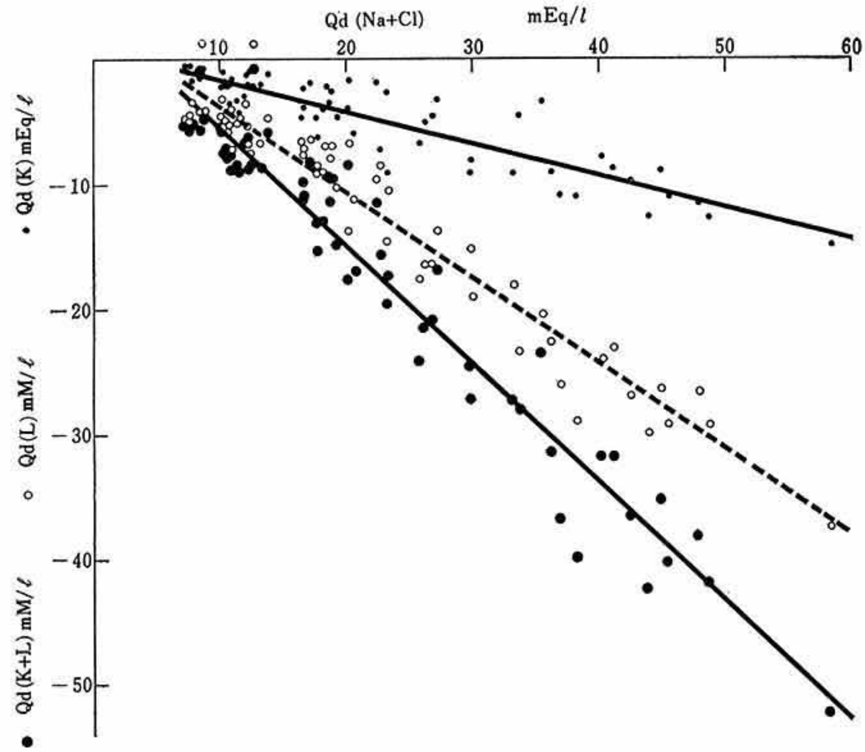

Fig. 2. Interrelationships between the increase of Na+Cl[Qd(Na+Cl)], and the decrease of K[Qd(K)], lactose [Qd(L)] and K+L[Qd(K+L)] in the abnormal quarter milk.

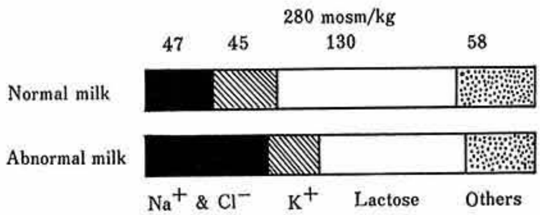

Fig. 3. Contribution of milk constituents to osmolarity of the normal and an abnormal quarter milk.

On the basis of the foregoing findings, the practicability of the estimation of increased Na^+ and Cl^- in abnormal milk by means of the electrical conductivity measurement was examined.

Detection of subclinical mastitis by the electrical conductivity measurement

Many attempts had been made for several years on application of the electrical conductivity test of milk for detection of subclinical mastitis. The electrical conductivity (EC) of milk is determined largely by the concentration of Na^+, K^+ and Cl^- in milk. But it is only few years since the measurement of milk electrolytes has become popular in laboratories. Up to then the EC value of milk was compared either with the results of bacterial examinations or leucocyte counts and so forth. Therefore, it was difficult to establish a criterion for diagnosis by the EC test for the detection of abnormal milk due to subclinical mastitis on a firm theoretical basis.

Since around 1970, Linzell and Peaker in Cambridge studied extensively physiological aspects of the electrolytes in milk. They studied also the EC of milk with parallel measurements of Na, K, Cl and lactose using foremilk of cows and goat's milk for detection of subclinical mastitis[4,5]. They confirmed once again the necessity of the simultaneous comparison of the four milks from separate quarters of individual udder at a milking, which Davis had proposed in 1947, and showed that infected quarters were detectable by the EC method with a high efficacy. They used criteria, such as I.M.S. which was a statistical measure of departure from parallelism with which the EC value of healthy quarters of individual udder tended to move from day to day, and such as differential conductivity (the ratio of the highest EC value divided by the lowest) a statistic first proposed by Davis (1947)[6] and later adopted by Greatrix et al. (1968)[7].

In the present study, the author set up a criterion for detecting abnormal quarter milk by the EC measurement on the basis of the Qd(Na+Cl) method[8]. With 168 quarter milk samples taken by separate quarter milking from 8 cows, a close correlation, $r=+0.95$, between the sum of the electrolytes, Na+K+Cl (mEq/l) and the EC value was found. The conductivity was measured at 40°C and expressed in terms of the specific conductivity (10^{-4} mho/cm^2).

The Quarter difference of the Electrical Conductivity, Qd(EC), of milk is obtained in the same manner as Qd(Na+Cl); that is the difference between the lowest and the others in the four quaters of an udder. Qd(EC) value depends on the difference in the electrolyte content or Na+K+Cl of the two quarter milks. The correlation between the two Qds was $r=0.98$ $(n=144)$.

Since the Qd(EC) method for detecting abnormal milk has its theoretical basis on the Qd(Na+Cl) method, results of determination by both methods must be highly correlated each other. As a matter of fact, very high correlations, $r=0.96$ $(n=126)$ for quarter milk and $r=0.95$ $(n=108)$ for foremilk were obtained. The regression equations of them were; $y=(0.334\pm0.010)x-0.090$, for quarter milk, where x is Qd(Na+Cl) mEq/l, y is Qd(EC) 10^{-4} mho, and for foremilk; $y=(0.330\pm0.010)x-0.283$. The relationship was

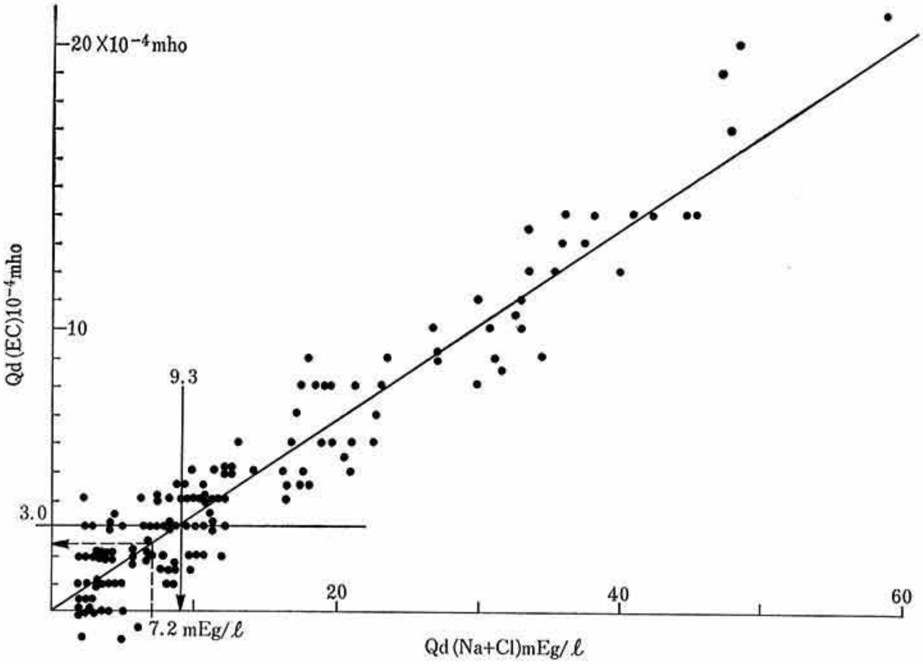

Fig. 4. Relationship between the quarter difference of Na+Cl value and that of the electrical conductivity of the quarter milks.

shown in Fig. 4.

As mentioned above, the criterion for Qd(Na+Cl) method was 7.2 mEq/l. Using the above regression equations the equivalent criterion for Qd(EC) method was calculated to be 2.3×10^{-4} mho of Qd(EC) against 7.2 mEq/l. Taking into account the fluctuation in reading the EC value, 3×10^{-4} mho was preferred to 2.3×10^{-4} mho for the practical use.

Comparison of the quarter difference of the electrical conductivity method with other methods

In Qd(EC) method, the quarter milk with conductivity higher by more than 3×10^{-4} mho (or 2.4×10^{-4} mho at 25°C) than the lowest quarter milk within an udder at a milking is graded abnormal. This Qd(EC) value corresponds to 9.3 mEq/l of Qd(Na+Cl)[8]. The quarter milk graded abnormal by each of the two methods using 1029 quarter milk samples from 25 cows was compared[9]. Twenty four percent of all milk samples was graded abnormal by the both methods and 72% non-abnormal in the same way; therefore the overall agreement was 96%.

The relation between Qd(EC) value and California Mastitis Test (CMT) score was as the following. The correlation of CMT score and Qd(EC) values of 1113 quater milks taken repeatedly from 123 quarters of 31 cows is shown in Table 1. Distribution of Qd(EC) values in each CMT class has a wide range.

Table 1. Correlation between the quarter difference of the electrical conductivity, Qd(EC), and California Mastitis Test

CMT score*		−				±				+				++				+++			
Group of quarter milk**		A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D
	0	288	26	10	3	9	49	6	3	2	18				1						1
(−)	1	124	14	19	6	7	16	10	7	1	8	6	6								2
	2	72	15	25	3	6	11	8	16	1	10	22	11			2	4			1	
	3	12	5	21	13	2	4	6	17		4	5	10			1	4			2	
	4	1	1	1	2			6	8				10			3	6			1	3
	5				3			2	9			1	5			1	4				4
	6				4				2				3				2				2
Qd(EC)	7				3				1				2				2				3
10^{-4} mho	8				2				4				3				1				2
	9				2				4				3				1				4
(+)	10								1				2				1				4
	11				1								1								1
	12																1				4
	13																				5
	14																				3
	15																				2
	15<																				11
Agreement %		97	90	71	29	8	5	37	64	0	10	18	70		0	71	85			75	94
		89 (605/676)				31 (66/214)				37 (49/134)				79 (27/34)				93 (51/55)			
		Overall 71 (788/1113)																			

* CMT score was graded according to PL-Tester (Nippon Zenyaku Kogyo Co., Ltd.)

** See text.

In the quarter milks (n=676) with CMT score of (−), 89% of their Qd(EC) value was smaller than 3×10^{-4} mho or (−). In the milks with CMT scores of (++) or (+++), 79 and 93% of Qd(EC) values were larger than 3×10^{-4} mho or (+), respectively. Agreements were fairy good in these classes. On the other hand, in the classes with CMT scores of (±) or (+), the agreement was low; 31 and 37%, respectively.

The same samples were classified further into 4 groups from A to D in the Table 1 according to degree of presumed damage or trouble in a quarter by referring to their Qd(Na+Cl) values, and the relation between CMT score and Qd(EC) value was reexamined[9]. As a result it was known that such Qd(EC) values as larger than 6×10^{-4} mho appeared exclusively in the quarter group of a high degree of damage (group D), and agreement of the two methods varied with the group to which the milk sample belonged. In the quarter groups with a little or probably no damage (group B and A), over 90% of Qd(EC) values of milks were (−) when CMT scores were (−) or (±). On the other hand, in the groups of relatively high damage (group C and D), Qd(EC) values tended to be (+), notwithstanding their CMT scores were (−) or (±).

Conclusion

Detection of abnormal quarter milk by the quarter difference in the electrical conductivity method depends on the same principle as in the case of quarter difference in the Na+Cl value method. The principle is based on the mechanism of formation of abnormal milk which appears frequently in some fixed quarter or quarters of individual udder of cows. In a word, abnormal quarter milk is formed by an admixture of 'true' milk with extracellular fluid or exudate within a mammary gland[10]. And, Qd(Na+Cl) value of an abnormal milk is considered as an index of the amount of exudate mixed in the milk[8]. Therefore, the amount of exudate in an abnormal milk and at the same time the extent of damage in a quarter are measured by the Qd(EC) method.

There are various indirect tests for detecting abnormal milk or subclinical mastitis. Although results of these tests are roughly correlated each other, there are certain discrepancies among them because each method examines different properties of milk. For instance, CMT examines the number of somatic cells in milk, but the electrical conductivity has no direct relation with the number of the cells. Therefore, combinations of indirect tests have been practised for the purpose.

The quarter difference in the electrical conductivity is one of the easiest means for detecting abnormal milk. In addition, there is a further possibility that abnormal quarter milk could be detected automatically during each milking time if the conductivity sensors are incorporated properly in the milking machine. If the idea is materialized, labor and time for milk sampling from each quarter of cows which is, at present, a great trouble in testing subclinical mastitis could be saved.

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