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Factors Affecting Milk Freezing Point from Montbeliard and Prim Holstein Cows

Data comprised 10,000 milk samples from 165 Holstein-Montbeliard cows in three geographical separated zones, geographically located in the ALGHARB region of Morocco. Milk freezing point (MFP), Temperature and precipitations were measured for a period from January 2015 to December 2016. The objective of this study was to estimate the influence of climatic parameters (temperature, precipitations), breed, month of milk sampling on the milk freezing point of Holstein-Montbeliard cows. The milk freezing points of samples were highly significantly affected by two factors: month of sampling and climatic parameters, specially temperature. MFP was lowest in milk samples taken from January to March, and those taken in summer period, highest in samples from November to December.

Keywords: *Freezing point / Raw milk / Seasonal / cow / climatic / rainfall*

1. INTRODUCTION

Milk is produced by a part of the cow called the mammary gland. (All mammalian species use mammary glands to produce milk.) One of the ways that the mammary gland works is to produce milk with the same overall "osmotic pressure" as the animal's blood has. Osmotic pressure is basically a measure of how much stuff is dissolved in a liquid, which determines the liquid's freezing point. Because of the important role of blood in the body, the body works very hard to keep its osmotic pressure at an ideal level, and that doesn't vary much from animal to animal, all cows have basically the same physiological requirements for their blood.

An interesting way that the milk freezing point is used is to determine if milk is watered down - either intentionally by the producer or naturally as a result of the cow being unhealthy. The osmotic pressure, and hence the freezing point of cows' blood remains fairly constant. The cow has to balance the osmotic pressure of its milk with that of its blood hence the milk freezing point, as a biological, is maintained fairly constant.

The Milk freezing point is the constant physical-chemical property of milk which is determined only by its water-soluble components such as lactose and salts, which in accordance with the Wigner law are held in milk at an approximately constant concentration.

Several studies have reported that the milk freezing point is influenced by factors related to variation in environment, management and breed [1,2].

Buchberger mentioned wider and deeper spectrum of factors: stage of lactation, age, health status, breed, milk yield, feed quality and amount, season, region, milking time and others [3], other researches show that not only extraneous water presence affects the MFP but there are other factors such: milk constituents, dairy cow breed, stage of lactation, dairy cow's nutrition, water intake, climatic conditions, regional and seasonal variations [3-4-5].

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2. MATERIALS AND METHODS

2.1 Data

One litre bulk of milk was made by mixing the morning and the evening milk and stored at 5°C. The cows were milked by hand in milking boxes during feeding, samplings are taken on a daily basis and the measurement of freezing point is made in a laboratory, meteorological measurements (temperature and precipitation rate) are carried out by pluviometer for precipitation rate and thermo-hygrometer for temperature.

The study variables are indexed according to a symbolization Xa,b for the milk freezing point, Ya and Za for expressing successive precipitation and average temperature data.

Table 1. Symobolization for the study parameters

Digital index	Zone	Race	Freezing Point	Precipitation	Temperature
	a	b	X	Y	Z
1	Sidi Slimane	Montbeliard			
2	Kenitra	Holstein			
3	Sidi Kacem				

2.2 Test Population data

The tested population is selected from three farms located in Kenitra, Sidi Kacem and Sidi Slimane, the choice is justified by the quality and vigilance of livestock management.

Table 2. Distribution of test population according to breed and farms

Farm	Test Population		Age (month)	
	MT	HT	Interval	Average
Sidi Kacem	54	0	[33-52]	35
Sidi Slimane	38	24	[31-50]	37
Kenitra	33	16	[31-55]	33

MT : Montbeliard
HT : Holstein

To eliminate the feeding effect, the feeding beha–vior of the test population cows for the three farms follows a standard diet as shown in the following table:

Table3. Feeding behavior on winter and Pasture

	Forage	Concentrated
Autumn	Silage herb	15% MS of the ration
Winter	Corn silage	30% MS of the ration
Spring	Grass grazed	1 KG
Summer	Grass grazed	0 KG

2.3 Laboratory Analysis and climatic control parameters

For the purpose of the study, each farm has identified the population cows for isolation, and the isolation makes it possible to properly manage the food system (quantity of food, quantity of water), as well as periodic veterinary checks. The disease of a cow of the population elimi–nates it from taking raw milk until its recovery.

Each day, the average test population for each farm is measured, and samples are sent to the laboratory for raw milk freezing point analysis, meteorological measureme–nts (temperature and precipitation rate) are carried out by pluviometer for precipitation rate and thermo-hygrome–ter for temperature. The results of analyses are collected after 30 hours, the freezing point is an indirect measure of the osmotic pressure [6], measured by thermistor cryos–cope, using a cryoscopy method [7], the measures of free–zing temperatures are given in absolute value (in |m°C|).

The results of analyses were measured for three representative samples of each delivery of raw milk. The milk freezing point is slightly less than that of water, because the presence of dissolved solids lowers the freezing point, it can range from -0.53°C to -0.575°C with an average of -0.555 °C [8].

3. RESULTS

3.1 Methodology for statistical analysis of results

Of 10,000 raw milk samples were collected from all three farms over a period of 24 months, the analysis of the results which allowed detecting about 678 out–liers, with 745 samples without results (for–getting analysis, forget–ing of sampling, samples of the non-conforming condition of transport, national holidays ...). While 8577 samples are considered compliant for statistical analysis, 85,7% of the collected samples constitute the statistical analysis database whose the results are summarized in Table 4.

3.2 Statistical Analysis

The statistical processing of the data makes it possible to establish links between the variables and to select the climatic parameters which have a significant influ-ence on the characteristics of raw milk, average liters and physicochemical composition of raw milk.

The analysis of the results is carried out by the pro-fessional MINITAB statistical data processing software (version 16.1.0.0).

4. DISCUSSIONS

In different countries, where many factors are different, the freezing point is variable. For example in Morocco, Bouisfi found that his average is -0.521 °C [9], in Latvia the milk freezing point ranged from -0.640°C till -0.494°C [4], in Germany from -0.531°C till -0.468°C [3], in Poland -0.540°C and -0.570°C [10], in Estonia -0.550 °C and -0.497 °C [1], American researchers found that it ranged from -0.550°C till -0.512°C [11]. The mean milk freezing point in Italy is -0.528°C [12], in Netherlands -0.521°C [13], in Czech Republic -0.523°C [14], in Switzerland -0.526°C [15], in the UK -0.539°C is different too [16].

The mathematical relationship between the freezing point depression and concentration of solute was wor–ked out by Raoult [17] and is expressed in the following formula :

$$T_f = K_f\, m$$

where T_f is the freezing point depression, K_f the molal depression constant, and m the molal concentration of the solute. The molal depression constant differs for each solvent. In water, it is 1.86 ° C. As Raoult pointed out, this exact relationship is valid only for dilute solutions of undissociated solutes.

4.1 Interval plot Milk Freezing Point 2015-2016

The freezing point oscillated between 511.38 |m°C| and 529.67 |m°C| in 2015, between 513.57 |m°C| and 528.95 |m°C| in 2016 and highly dependent on the season, the average deviation of the freezing point between the mi–nimum and maximum freezing point of each pair (zone, breed) is of the order of 11.24 |m°C| in 2015 X(1,1), 6,88 |m°C| In 2016 X(2,2), this difference is justified by the effect of thermal stress, due to the extreme temperatures recorded in zone 1 (T° Max reached 36.9°C).

Brâthen and Mitchell are confirmed that changes in temperature and diet are considered to be primarily responsible for the seasonal effect on MFP [18,19] , the exposure of the herds to extreme temperatures is re-lated to the duration of the day, i.e. the duration of ex-perimental illumination.

The MFP interval is approximately the same for both breeds, the difference is seen between the experi-mental zones. The climatic conditions especially the temperature explain these observations observed in the two years, milk freezing point is influenced by factors related to variation in environment, management, and breed [1,2]. The daily freezing point monitoring shows that in every zone, the pace of the freezing point varia-tion is identical and does not vary between breeds. This observation may be justified by the rearing environ-ment, the same effect of geographic region on MFP was found by Kedzierska [20].

Table 4. Results of raw milk freezing point and Meteorological Characteristics of zones

Months	Freezing Point (|m°C|)					Temperature °C									Precipitations (ml)		
Zone	SK	SS		KN		SK			SS			KEN			SK	SS	KN
Breed	MT	MT	HT	MT	HT	MX	MIN	AVR	MX	MIN	AVR	MX	MIN	AVR			
Jan-15	515.7	524	516	528.7	525.7	18.2	7.1	12.7	17.1	6.1	11.6	17.7	7.9	12.8	116	124	122
Féb-15	516.8	521	523.2	527.3	523.1	18.4	7.0	12.7	17.5	6.2	11.8	17.8	7.7	12.7	47	48	36
Mar-15	516.1	516.6	521.1	526.1	522.7	21.4	8.4	15.0	20.8	9.8	15.3	20.3	8.4	14.4	29	32	28
Apr-15	513.4	516.5	518	523.6	522.7	25.9	11.4	18.7	24.2	10.2	17.2	24.0	11.3	17.6	49	50	50
May-15	512.5	515.9	516.9	522.9	522.9	31.8	14.1	22.6	26.5	10.6	18.6	24.4	13.8	19.0	03	00	15
Jun-15	512.3	514.4	516.1	526.1	528.4	31.3	16.3	23.8	32.6	14.8	23.7	28.1	16.0	22.1	00	00	00
Jul-15	517.1	516.6	518.8	527.6	529.7	32.7	18.0	25.4	35.6	15.5	27.1	28.4	17.5	22.9	00	00	00
Aug-15	519.4	518.6	518.2	527.9	527.3	35.0	18.8	26.9	36.9	19.0	28.0	30.1	18.0	24.1	00	00	00
Sep-15	515.7	514	517.5	525.2	525.2	31.7	18.3	25.0	31.5	18.4	25.0	28.5	17.5	22.3	7.3	14	6.6
Oct-15	515	513.4	516.1	524.3	523.1	31.2	15.9	23.6	28.5	13.8	21.2	29.3	15.7	22.5	29	16	52
Nov-15	512	513.2	515.4	521.9	521.8	22.8	12.4	17.2	21.7	7.9	14.8	21.2	12.4	16.8	195	271	299
Déc-15	511.4	512.8	515.3	521.9	521.8	17.1	7.8	12.5	19.2	3.9	11.6	16.8	8.1	12.5	75	112	116
Jan-16	517.9	519.7	519.5	528.8	525.8	17.0	8.9	10.1	16.9	6.5	11.2	17.6	7.8	12.7	144.9	66.1	113
Féb-16	520.2	519.5	520.4	528.8	527.5	16.9	6.1	11.2	18.1	7.0	12.0	17.4	6.6	12.0	49.1	43.9	30.4
Mar-16	518.4	520.4	519	527.4	525.9	19.4	10.4	14.6	20.7	8.2	13.7	19.6	11.0	15.3	185.7	125	143
Apr-16	517.3	518.4	518.6	525.5	525.5	23.8	11.2	17.2	21.8	9.6	15.8	22.7	11.1	16.9	37.1	43.3	61.0
May-16	516.7	518	518	524.5	525.4	25.5	11.8	18.3	22.9	9.4	19.1	23.9	11.8	17.9	23.6	30.4	25.0
Jun-16	516.1	517.2	518.3	526.9	524.7	32.1	15.9	23.6	28.4	10.8	21.2	28.3	15.5	21.9	00	00	00
Jul-16	518.8	517.7	519.3	527.5	525.7	35.6	19.4	26.9	29.6	12.1	25.4	30.1	18.7	24.4	00	00	00
Aug-16	519.5	519.1	519.1	528.9	527.7	37.4	20.0	28.7	30.4	15.6	25.2	32.1	19.5	25.8	00	00	00
Sep-16	517.1	518.1	519.2	525.8	525	32.1	19.4	25.7	25.6	19.2	22.6	29.6	18.6	24.0	7.5	8.5	21.6
Oct-16	514.9	518.4	517.9	525.6	523.7	28.9	14.9	21.9	24.3	17.3	20.2	26.5	14.8	20.7	15.5	4.2	25.7
Nov-16	513.6	516.9	515	524.7	523.5	21.2	8.15	14.8	21.5	10.1	13.1	21.3	8.9	15.1	89.6	62.2	61.2
Déc-16	514	516.7	517.8	522.1	520.9	20.3	5.3	13.0	20.1	6.5	12.0	18.7	6.5	12.6	43.5	48.1	47.8

SS : Sidi Slimane - SK : Sidi Kacem - KN : Ken

It is noted that during the two years of testing; the pace is the same at zone 1 for both breeds; the same is also true for zone 2, for the zone 3 with a single breed follows a different pace from other areas, according to these results the breed has not a significant influence on the freezing point variability, who can be explained by the differences between milk constituents, the same observations cited by Agnieszka [21].

Of the factors examined by M. Heno, Maximum FP differences for each of the factors studied were as follows: 0.00221°C for breed, 0.00321°C for lactation stage, 0.00531°C for interaction of year and period. Interpreting the milk freezing point data, primarily the possible effects of season and diet, and the combined effect of them should be considered [1].

4.2. Follow-up Freezing Point 2015-2016

Milk freezes at a lower temperature than the water. The average value of the milk freezing point measured on a cryscope is generally 540 (-0.540 ° C). Pure water must have a freezing point of 0 °C. The addition of water to the milk modifies the freezing point of the latter to make it rise towards that of the water. The milk free–zing point is a "physiological constant". This does not mean that it is invariable. In fact, diet, breeding technique, season, climate, time of lactation, all has a significant effect on a given sample.

MFP was lowest for milk sampled from January to March and during the summer period (July and august), and highest at the end of the year (November and December). Our study indicates a highly significant effect of the month of milk sampling on the milk freezing point, the mean value of MFP was observed during the end of summer (September) and the first month of spring (April). The effect found for season of milk sampling on MFP varies among studies. Our study indicates that MFP was higher in winter than in the summer, reversing the finding of Bjerg and Henno, whose reported that MFP was higher in summer than in winter [1-22].

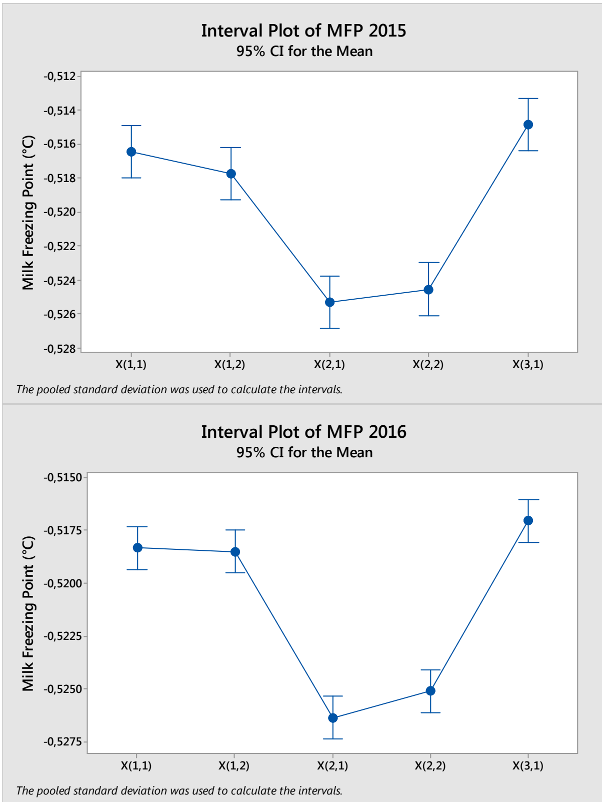

Fig 1: Interval plot of freezing point 2015-2016

Bjerg M. and Henno M. suggested that changes in temperature and diet were mainly responsible for the seasonal effect on MFP. Janstova found that the milk freezing point of goat was lowest in summer and explained that organism dehydration during hot summer days could reduce MFP [23]. Brzozowski and Zdziarski reported that MFP was higher in the autumn-winter period and lower in the pasture season.

Additionally, they found significant effect of month of year on fat and protein content of milk [24]. Kedzierska-Matysek did not fnd a significant effect of season on the MFP of Polish cows. They found that the milk obtained in the autumn-winter season had a more favorable chemical composition, including the concentrations of fat and protein, as compared to the milk obtained in spring-summer [20].

The relatively big difference between MFP occurring in adjacent months of the winter season (December and January) could be partly explained by the results of Brzozowski and Kedzierska - Matysek who

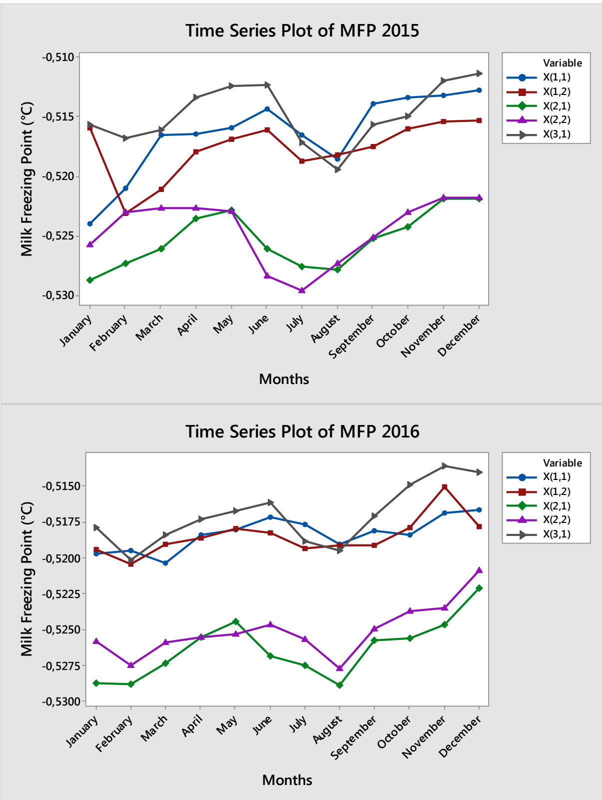

Fig 2: Times series plot of freezing point 2015-2016

expressed that the depression of the milk freezing point was often related to an increase in protein and solids content as well as to a decrease in the lactose percent in milk [20-25].

The variation of the freezing point was expressed as a function of time, the graphs on figure 2 present the results obtained. MFP decreased during summer compared with winter, caused by increased water intake due to increased temperature and sunshine hours, the same conclusion expressed by Bjerg and Brzozowski [22-24].

With a standardized feeding regime, the tests carried out showed that the breed did not affect freezing point evolution and since the diet is standardized for the three zones, it can be concluded that climatic conditions and average temperature have an impact on the variability observed at the milk freezing point. These results are supported by the results of Buchanan and Lowman, they observed changes in freezing points with the seasons. They concluded that these changes were due primarily to changes in feed. However, they stated that meteorological conditions may have been a contributing factor [26].

Regan and Richardson observed that changes in environmental temperature affected the freezing point [27]. Aschaffenburg and Veinoglou suggested that differences in environmental temperature were a contributing factor to the seasonal variations that they observed [28].

5. CONCLUSION

The objective of this study was to investigate the climatic and seasonal influence on the milk freezing point. MFP was lowest for milk sampled from January to March and during the summer period (July and august), and highest at the end of the year (November and December). The average deviation of the freezing point between the minimum and maximum freezing point of each pair (zone, breed) is justified by the effect of thermal stress. The exposure of the herds to extreme temperatures is related to the duration of the day, i.e. the duration of experimental illumination.

The results show that temperature changes and season had the greatest effect on MFP, where maxi–mum deviations were showed in summer period, espe–cially August, caused an increase of water intake due to increased temperature and sunshine hours. The MFP as one of the physical characteristic, including viscosity of rheological material (raw milk) are stron–gly dependent on their microscopic and macroscopic struc–tures, as well as the presence of intermolecular forces [29], the pace of the freezing point variation is identical and does not vary between breeds. This observation may be justified by the rearing environment, the combined effects of all the above-mentioned factors may result in a significant (P 0.05) increase in MFP.

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AUTHOR CONTRIBUTIONS

Conceived and designed the experiments: M.Chaoui.
Performed the experiments: A.Bouisfi,
Analyzed the data: F.Bouisfi, A.Bouisfi, M.Chaoui.
Contribued materials/analysis tools: M.Chaoui.
Wrote the paper: A.bouisfi.

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ФАКТОРИ КОЈИ УТИЧУ НА ТАЧКУ МРЖЊЕЊА МЛЕКА КРАВА РАСЕ ХОЛШТАЈН И МОНТБЕЛИАРД

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Подаци обухватају 10 000 узорака млека узетих од 165 крава расе Холштајн-Монтбелиард из три региона географски лоцираних у обасти Алгарб у Мароку. Тачка мржњења млека, температура и

падавине су мерени у периоду јануар 2015 – децембар 2016. Циљ истраживања је био да се утврди утицај климатских параметара (температуре и падавина), расе и месеца узорковања на тачку мржњења млека узетог од крава расе Холштајн-Монтбелиард. Утврђен је високо сигнификантан утицај два фактора на тачку мржњења млека: месец узорковања и температура. Најнижа тачка мржњења је била код узорака млека узетих у периоду јануар-март и током летњег периода, а највиша је била у периоду новембар-децембар.