

## A METHOD TO DETERMINE CUT-OFF VALUES IN MILK SOMATIC CELL COUNTS TO CHARACTERIZE UDDER HEALTH OF DAIRY COWS USING TEST DAY RESULTS

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*Dans la plupart des pays, le seul critère disponible en routine décrivant le statut sanitaire des troupeaux bovins laitiers vis-à-vis des infections mammaires est la numération cellulaire individuelle (NC) du lait des traites de 24 h estimée à chaque contrôle laitier. L'objectif de cette étude est de présenter une méthode de détermination de valeurs seuils de NC permettant de classer les vaches selon leur statut vis-à-vis des infections mammaires. Les données (60 208 enregistrements) de 6 502 vaches laitières dans 105 troupeaux Holstein de Loire-Atlantique (France) ont été utilisées. La médiane, les premier et troisième quartiles des NC étaient respectivement 85 000, 38 000, 193 000 cellules/ml. Le principe de détermination des valeurs seuils de NC a été le suivant : (1) la distribution de NC résultait de l'addition de n sous-populations de NC distribuées normalement  $N_i (m_i, \sigma_i^2)$  et d'effectif  $k_i$ , avec  $i$  de 1 à  $n$ ,  $m_{i+1} > m_i$ , et  $\sigma_{i+1}^2 > \sigma_i^2$ . La meilleure combinaison ( $n, (m_i, \sigma_i^2, k_i)$  pour  $i$  variant de 1 à  $n$ ) était celle qui minimisait la valeur du  $\chi^2$  résultant du test de comparaison des distributions de NC observée et théorique (addition des n sous-populations) ; (2) les valeurs seuils de NC séparant 2 sous-populations  $i$  et  $i+1$  étaient celles qui maximisaient le produit des valeurs prédictives positive et négative pour une valeur de NC donnée d'appartenir à l'une ou l'autre sous-population. Quatre sous-populations ont été retenues. Les valeurs seuils de NC sont 45 000, 155 000 et 235 000 pour les primipares et 35 000, 175 000 et 325 000 chez les multipares. Ces seuils peuvent être utilisés pour décrire (1) le statut sanitaire des vaches laitières vis-à-vis des infections mammaires à un contrôle donné, et (2) la dynamique des infections mammaires aux niveaux vache et troupeau.*

### INTRODUCTION

In dairy herds of most countries, the only information describing udder health and routinely available consists in bulk milk and/or individual Somatic Cell Counts (SCC) measured at successive test days within the lactation. To characterize udder health of the herd, a preliminary step is to classify cows according to their presumed udder health status. This paper aimed at answering this step, that is to present a method to determine possible SCC cut-off values to classify cows based on their individual SCC test day results.

### MATERIAL AND METHODS

105 dairy farms were randomly selected from those enrolled in the milk recording scheme (Loire-Atlantique area, France) between 1994 and 1995. At each test day, individual SCC (x 1,000 cells/ml) were recorded for each lactating cow (milking of the 4 udder quarters within a 24 hours period). A total of 62,493 records were considered during the study period.

Records with SCC over 1,200,000 cells/ml were excluded from the analysis, assuming that SCC over 1,200,000 cells/ml were associated with an obvious poor udder health status. The resulting dataset consisted in 60,208 records from 6,502 Holstein cows. Records from primiparous cows accounted for 33.1% of the data. Descriptive criteria of SCC for primiparous and multiparous cows are displayed in Table I.

**Table I**  
**Descriptive criteria of individual SCC for primiparous and multiparous cows**

	Individual SCC (x 1,000)				
	Mean	Sd	Percentile25	Percentile50	Percentile75
All cows-records (n = 60,208)	161	201	38	85	193
Primiparous cows-records (n = 19,945)	128	161	36	72	147
Multiparous cows-records (n = 40,263)	178	217	40	94	220

SCC were categorized in classes (range: 10,000 cells/ml each). The distribution of SCC was right kurtosed both in primiparous and multiparous cows. The observed distribution of SCC was assumed to fit a theoretical distribution resulting from the addition of n subpopulations normally distributed  $N_i (m_i, \sigma_i^2)$  of SCC, with  $i$  from 1 to  $n$ ,  $m_{i+1} > m_i$ ,  $\sigma_{i+1}^2 > \sigma_i^2$ . Each population  $i$  included a number of observations  $k_i$ , and was left truncated at SCC equal to 0. The resulting theoretical distribution varied according to the number of subpopulations ( $n$ ), the mean, the variance and the number of observations of each subpopulation ( $m_i, \sigma_i^2, k_i$ , respectively).

The assessment of the (n-1) SCC cut-off values was performed in 2 steps.

The first step aimed to provide the best combination of the n  $N_i (m_i, \sigma_i^2)$  and  $k_i$  minimizing the chi-square value from the comparison test of the observed and theoretical distributions (goodness-of-fit criterion). For a given subpopulation  $N_i$ , 3 parameters ( $m_i, \sigma_i^2, k_i$ , respectively) must be assessed. When considering n subpopulations, the assessment of the best combination of the n  $N_i$  (objective to be reached) necessitated to investigate a large range of values of  $m_i, \sigma_i^2, k_i$  for a given  $N_i$ . Two different programs were used in that purpose. In order to optimize

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computing time and to avoid the investigation of a priori non suitable combinations, a spreadsheet program was built to provide ranges of possible values of  $m_i$ ,  $\sigma_i^2$ ,  $k_i$  based on visually assessed agreements between the theoretical and the observed distributions. Combinations of range of values deemed to be possible from the spreadsheet program were then loaded in a program written in the Microsoft Access™ Basic language 2.0 for mathematical optimization. Various numbers (2 to 5) of subpopulations  $n$  were investigated, and the assessment of relevance to consider an additional population  $i+1$  to the  $n$  initially investigated was based on the minimization of the chi-square value from the comparison test of the observed and theoretical distributions resulting from the addition of  $n$  or  $n+1$  subpopulations respectively over the same range of SCC.

In a second step, once the best combination of  $n$ ,  $m_i$ ,  $\sigma_i^2$ ,  $k_i$  determined, the SCC cut-off value between 2 consecutive subpopulations  $i$  and  $i+1$  was assessed using the positive predictive value and the negative predictive value for a SCC to belong to subpopulation  $i$  or subpopulation  $i+1$ , in case of bimodal distribution (Martin et al., 1987). Retained SCC cut-off value maximized the product of the positive predictive value and the negative predictive value.

This procedure was used for primiparous and multiparous cows separately.

## RESULTS

For both primiparous and multiparous cows, the observed distribution of SCC resulted from the addition of 4 normally distributed subpopulations. The best combination of the  $n$ ,  $m_i$ ,  $\sigma_i^2$ ,  $k_i$  (for  $i = 1$  to  $n$ ) was assessed for SCC over the range 10 000 to percentile 95 (441,000 and 674,000 cells/ml in primiparous and multiparous cows respectively), because SCC distribution was almost uniform over the range (percentile 95 - maximum). Table II provides statistics of the 4 subpopulations retained from the analyses.

**Table II**  
**Descriptive statistics of the 4 subpopulations retained from the analysis (SCC x 1,000)**

Subpopulations	Subpopulation size		Mean		Sd	
	Primiparous	Multiparous	Primiparous	Multiparous	Primiparous	Multiparous
Subpopulation 1	4887	8455	28	15	13	21
Subpopulation 2	9374	18118	62	58	38	53
Subpopulation 3	2592	7247	160	198	45	75
Subpopulation 4	3092	6443	260	405	125	180

Table III provides the predictive positive values, negative predictive values and cut-off values for a given pair of 2 consecutive subpopulations for primiparous and multiparous cows. Figures 1 and 2 display the distributions of the 4 subpopulations, the resulting theoretical and the observed distributions for primiparous and multiparous cows respectively.

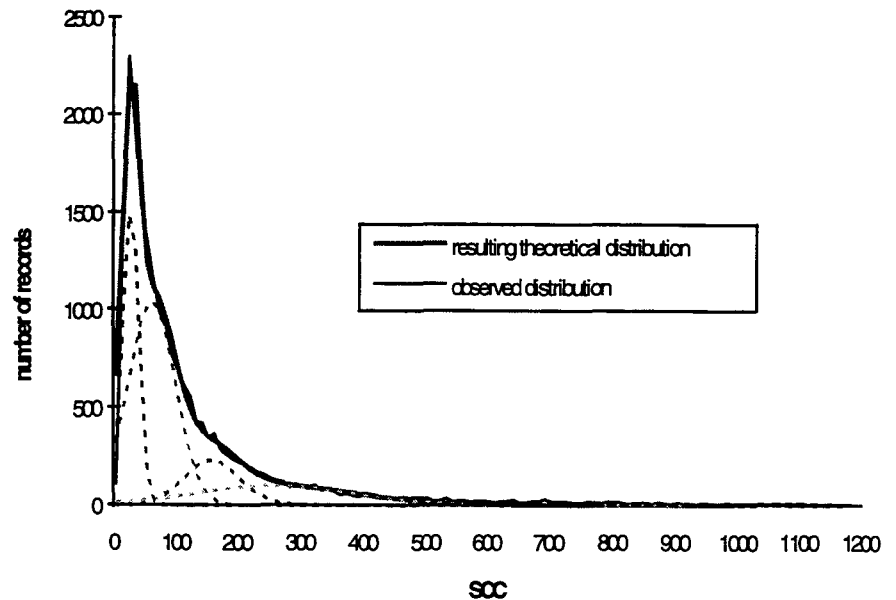
**Table III**  
**Cut-off values between the 4 subpopulations**  
**and corresponding positive and negative predictive values (SCC x 1,000)**

Consecutive subpopulations	Cut-off value		Positive predictive value		Negative predictive value	
	Primiparous	Multiparous	Primiparous	Multiparous	Primiparous	Multiparous
Subpopulations 1 - 2	45	35	0.97	0.91	0.59	0.60
Subpopulations 2 - 3	155	175	0.96	0.95	0.88	0.86
Subpopulations 3 - 4	235	325	0.94	0.95	0.65	0.76

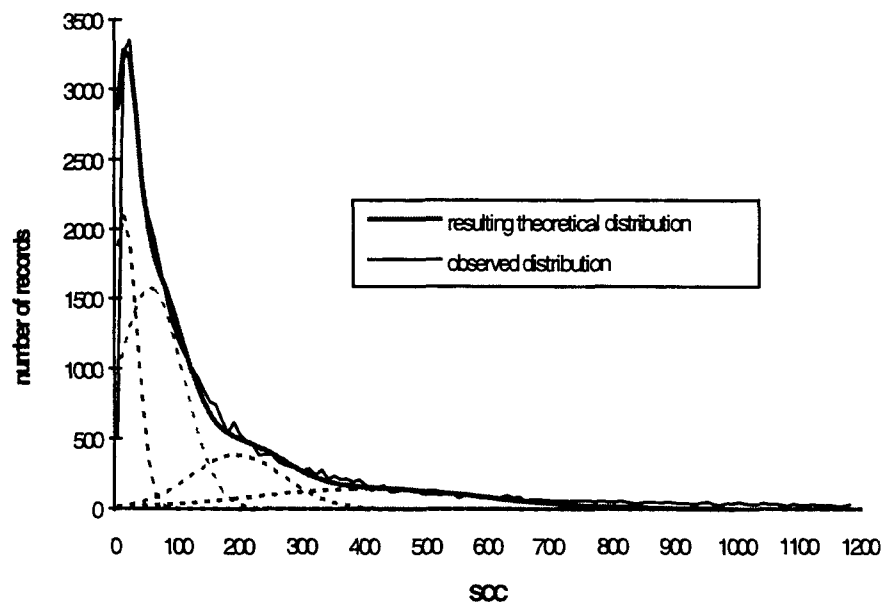
## DISCUSSION AND CONCLUSIONS

Both in primiparous and multiparous cows, individual test day SCC results may be classified in 4 classes. Using this method, it is then possible to classify cows according to their presumed udder health status. This study suggests at least three subpopulations within the range 0 - 300,000 cells/ml. Cut-off values are higher in multiparous than in primiparous cows (except for the first one), in agreement with descriptive statistics (Table 1). Whatever the lactation number, the first class (below 40,000 cells/ml) may include cows free from udder infection. Whether the 3 other described subpopulations fit populations infected by minor pathogens, and/or infected with major pathogens is questionable. In a study taking into account relationships between SCC and presence of minor or major pathogens in milk to determine SCC cut-off values (Seryes, 1985a), the reported cut-off values were different from the present ones. In addition our findings do not fit the mean SCC or SCC ranges based on infection status (absence of bacteria, presence of minor, major pathogens) described by Reneau (1986), Seryes (1985b), Schukken et al. (1989). The SCC values by infection status found in these latter studies largely differed. Furthermore Dietz et al. (1997) reported associations between certain alleles of the BoLA genes complex and elevations of SCC. Further research is therefore needed to investigate the assumption that the subpopulations described in the present study consist respectively in different populations based on interactions between cow genome and infection status. Furthermore, many other factors (e.g. lactation stage, milk yield at test day) are known to induce SCC variation. Stratified analysis are therefore suitable in further analysis to assess thresholds. These results may be of interest to characterize (i) herd udder health status at a given test day (see Thurmond, 1986), and (ii) dynamics of udder health status within the lactation at the cow level.

**Figure 1 :**  
Observed and resulting  
theoretical distributions  
of SCC in primiparous cows  
(SCC x 1,000)



**Figure 2 :**  
Observed and resulting  
theoretical distributions  
of SCC in multiparous cows  
(SCC x 1,000)



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