

## Evaluation of finger imprint persistence as a practical method for measuring the severity of mammary oedema in dairy cows

### Az ujjlenyomat tartósságának, mint a tejelő tehenek tőgyödéma súlyosságának mérésére szolgáló gyakorlati módszer értékelése

Violetta TÓTH<sup>1</sup>, László GULYÁS<sup>2</sup>, Edit MIKÓ<sup>3</sup> (✉), András GÁSPÁRDY<sup>4</sup>

<sup>1</sup> István Széchenyi University, Kázmér Albert Faculty of Mosonmagyaróvár, Antal Wittmann Multidisciplinary Doctoral School of Plant, Animal and Food Sciences, 9200 Mosonmagyaróvár, Vár Square 2, Hungary

<sup>2</sup> István Széchenyi University, Kázmér Albert Faculty of Mosonmagyaróvár, Institute of Animal Sciences, 9200 Mosonmagyaróvár, Vár Square 2, Hungary

<sup>3</sup> University of Szeged, Faculty of Agriculture, Institute of Animal Sciences and Wildlife Management, 6800 Hódmezővásárhely, Andrassy Street 15, Hungary

<sup>4</sup> University of Veterinary Medicine Budapest, Institute of Animal Breeding, Nutrition and Laboratory Animal Science, 1078 Budapest, István Street 2, Hungary

✉ Corresponding author: [miko.edit@szte.hu](mailto:miko.edit@szte.hu)

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#### ABSTRACT

The study aims to assess the severity of udder oedema around the calving using a traditional scoring (0-3 point scale) and the firstly applied fingerprint test to measure skin firmness. Furthermore, it was investigated how the degree of udder oedema is influenced by the effects taken into account in the processing. The study was carried out on 62 animals diagnosed with udder oedema before calving at a typical large-scale dairy operation in Hungary. It was found that with more severe udder oedema, the first udder teats are thicker ( $P = 0.038$ ) and the udder skin surface is cooler ( $P = 0.007$ ), and the duration of the fingerprint is significantly longer ( $P < 0.001$ ). There is a strong positive correlation between oedema severity and fingerprint persistence ( $r = 0.66$ ,  $P < 0.05$ ). This suggests that the persistence of finger imprint is a reliable indicator of the severity of udder oedema.

**Keywords:** Holstein-Friesian, udder oedema, teat diameter, udder skin firmness

#### ÖSSZEFOGLALÁS

A kutatás célja az ellés körüli tőgy ödéma súlyosságának megállapítása hagyományos pontozásos (0-3 pontos skálán) és a bőr rugalmasságát mérő elsőként alkalmazott ujjlenyomat próbával. Továbbá, vizsgálatra került, hogy a tőgy ödéma mértékét hogyan befolyásolják a feldolgozás során figyelembe vett hatások. A vizsgálat egy jellegzetes magyarországi tehenészet 62 ellés előtt tőgy ödémával diagnosztizált egyedén valósult meg. Megállapítást nyert, hogy súlyosabb tőgy ödéma esetén az első tőgybimbók vastagabbak ( $P = 0.038$ ) és a tőgybőr felszíne hűvösebb ( $P = 0.007$ ), valamint az ujjlenyomat tartóssága lényegesen hosszabb ( $P < 0.001$ ). Az ödéma súlyossága és az ujjlenyomat tartóssága között szoros pozitív kapcsolat áll fenn ( $r = 0.66$ ,  $P < 0.05$ ). Ez alapján arra lehet következtetni, hogy az ujjlenyomat tartóssága a tőgyödéma súlyossági fokának megbízható indikátora.

**Kulcsszavak:** holstein-fríz, tőgyödéma, tőgybimbó átmérő, tőgybőr rugalmasság

## INTRODUCTION

Udder oedema is the accumulation of lymph in the interstitial space of the udder and surrounding tissues (Tucker et al., 1992; Kojouri et al., 2015). Genetic factors play an important role in its development (Van Dorp et al., 1998), but in addition, one-sided feeding during the last stage of pregnancy is a predisposing factor (Block, 1994). Furthermore, the occurrence of oedema is typical of breeds and individuals with high milk production (Gilbert and Schwark, 1992).

On physiological grounds, edema that develops moderately around parturition cannot be considered an infectious metabolic disorder (Waller et al., 2007). Sixty-six % of Holstein-Friesian cows have at least one history of oedematous condition (Morrison et al., 2018). Moreover, it was also observed that cows with a longer-than-average gestation period had a higher incidence of severe udder oedema (Malven et al., 1983). Hillerton (2022) found that cows with a higher-than-ideal condition score had a higher incidence of physiological udder oedema. Dentine and McDaniel (1983) reported that autumn calving cows had more severe oedema than winter and spring or summer calving cows. Oedematous condition usually resolves spontaneously within a few days after calving, but within 3 weeks at the latest (Jackson, 1996; Durna Corum et al., 2021). Pre-partum udder oedema was found to be more severe and more frequent in heifers than in older dairy cows. Emery et al. (1969) speculate that the higher incidence of udder oedema in heifers may be due to less developed vascular circulation.

Okkema and Grandin (2021) have reported that more severe udder oedema has a negative effect on the longevity of the cow, because the ligaments that suspend the udder can be damaged. Swollen, oedematous udder teats make it difficult to apply the milking bowls, and mechanical damage to the teats can contribute to mastitis or dermatitis. Although udder oedema is a common metabolic disorder in dairy herds that may be responsible for damage to the udder and teats (Moroni et al., 2018), and may lead to an increased risk of clinical mastitis (Compton et al., 2007; Zigo et al., 2019; Hisira et al., 2023).

Fernandes et al. (2022) did not find a correlation between udder oedema and udder infections in their study. Ramos (2020) observed that in post-calving pathological udder oedema, the udder volume increases, swells, becomes asymmetric, the udder skin becomes erythematous, its elasticity decreases, and the temperature of the udder increases. Morrow and Schmidt (1964) observed that cows with severe or multiple udder oedema had more loose udder suspension and laterally directed udder teats. As a result of chronic udder oedema, the udder teats gradually shorten and thicken and the cavity of the teat duct may narrow, making milking difficult (Medrano-Galarza et al., 2012). Gussmann et al. (2019) observed higher culling rates in severely udder-oedematous cows.

Since udder oedema can have a detrimental effect on udder suspension and herd life, it would be advisable to minimise udder oedema in dairy herds, which would bring significant economic benefits. Ducrocq (1994) emphasised that the structural condition of the udder (udder attachment, teat placement, suspensory ligaments) has a major influence on the productive life. Waage et al. (2001) found that udder oedema and oedematous condition of the teats are associated with clinical mastitis around calving. Poor manifestation of conformation traits: an udder with loose suspensory ligaments, an udder close to the ground, an irregular udder, or too thick, too thin teats make the cow more susceptible to mastitis (Tóth and Bak, 2001). Severe pre-calving or persistent udder oedema may be risk factors for difficult milking, hardening of the udder tissue, necrosis of the udder skin and teats, mastitis, and reduced milk yield (Cook, 1998).

A congestion due to higher hydrostatic pressure in the blood vessels and reduced venous outflow from the udder bud may be a prerequisite for oedema (McGavin and Zachary, 2009). In ultrasound studies, Stauffer et al. (2021) found that the oedematous condition is first relieved at the base of the teats, irrespective of the severity of udder oedema. No signs of oedema were observed in the teat body, which may be explained by the limited increase in the diameter of the teats and the

fact that the milk in the teat cistern provides internal pressure that can prevent oedema. Similarly, Kuchler (2011) reported the absence of oedema-like features on ultrasound examination of the teat wall.

In their study, Bowers et al. (2006) found that heifers milked before calving had a lower udder oedema score after calving than heifers that were not milked before calving. The effect of milking before calving on udder oedema is not yet clear and uniform. The presence and severity of mastoid oedema is determining by the widely used inspection method in dairy cattle. This involves subjective scoring the degree of oedema on a scale of different sizes. Dentine and McDaniel (1983) compiled the literature on the most used system in the past, which was ranging from 1 to 5. Tucker et al. (1992) introduced a 10-point scale (that takes also abdominal swelling into account) used in rating udder oedema. Later, a more accurate scheme was developed by Morrison et al. (2018), where cows were assigned a score of 0 to 3.

In human medicine and pet veterinary practice (Argenta et al., 2022) the severity of oedema of other forms is identified by use of the pitting test (godet's sign). Here, when pressure is applied with the second finger to the swollen area, an indentation will remain. Oedema is graded according to the depth of the indentation when the godet's sign is positive and the time it takes for the skin to return to normal (Berry, 2021). In human, the pitting test is widely used to identify and characterise oedema of lymphatic and venous origin (Sanderson et al., 2015). When physically examining the oedema, the doctor should describe the location, time and extent of the indentation to set up a treatment plan (Trayes et al., 2013). However, the descriptions of how to perform the pitting test and how to evaluate it vary widely. The differences are most noticeable in the amount and duration of pressure. When describing the amount of pressure, some describe it as "as forcefully as possible" (Brorson, 2012) or "firmly but without pain to the patient" (Lymphoedema Framework, 2006). In terms of duration, experience ranges from 5 to 60 seconds (Stanton et al., 2006; Sussman and Bates-Jensen, 2006; Muldoon, 2011).

Avais et al. (2020) studied udder oedema in dairy goats which was determined visually or by pressing the udder with their finger. If the skin returned to its original state after more than 3 seconds, the goat was considered to have udder oedema. The depth of the pit and the time of skin return were not measured. Kaiser et al. (2020) investigated post-farrowing mastitis in sows, which included the assessment of udder oedema. The test only covered whether the udder was oedematous or not. The udder was considered oedematous if the gland was dough-like to the touch and a pit remained in it when pressure was applied. Shahzad et al. (2011) assessed the presence of udder oedema in buffaloes, also by visual and palpation methods, but did not assess the strength of the pressure, the depth of the pit and the time of skin return. Although, for illustrative purposes, external pressure on the udder with pronounced "plastic" swelling by fingertips, maintained for a short period as an indentation, was known (Swett et al., 1938), to determine the extent of oedema has never been widely used in cattle.

This study aims to elaborate a continuous rating method (persistence of finger imprint) to quantitate oedema severity that is fast, efficient, cost-effective, and easily integrated into the daily milking routines at a typical Holstein-Friesian large-scale dairy farm in Hungary and supports the visual assessment system of Morrison et al. (2018). A further aim is to determine the relationship between the persistence of index finger imprint and the udder oedema scores and the other examined traits (like age at first calving, gestation length, body condition score, front teats length, rear teats length, front teats diameter, rear teats diameter, udder skin temperature, central ligament score) and variables (like parity group, semester of calving, test day, udder treatment, reproductive treatment) as well.

## MATERIAL AND METHODS

### *The livestock examined*

The presence and severity of udder oedema were investigated at a Holstein-Friesian large-scale dairy operation in the Southern Great Plain of Hungary. The

number of lactating cows on the farm was 615. Cows were milked in a double 20-parallel milking parlour three times a day. Its special feature is that an ADF (Automatic Dipping & Flushing) teat disinfection system is built into the milking bowls. The hygienic milking program in use is effective and briefly includes the following. Under a pre-milking teat-cleaning regime the teats are dipped in disinfectant and then wiped dry with a paper towel. Foremilk samples are then expressed on a quarter level to perform CMT (California Mastitis Test) and the milking machine is attached. At the end of the milking process, the cups are removed manually in the calving pen and automatically in the milking house. Finally, the teats are disinfected and cared for after milking.

The lactating cows are housed in free stall barns with lying boxes of straw bedding. Dry cows and cows about to calve are kept in deep litter stalls using straw.

#### **Data collection**

A total of 62 cows with mammary oedema were selected from those that had been standing for about two weeks immediately before calving. The presence and the severity of udder oedema was determined by physical examination of the udder using a four-point scale developed by Morrison et al. (2018). Changes in udder oedema severity were monitored weekly for each selected individual until week 8 postpartum. The severity of oedema is scored from 0 to 3, where 0 = no oedema, 1 = mild oedema, 2 = moderate oedema and 3 = severe oedema.

For cohort comparison, more variables were considered as an outcome variable on oedema, additionally. These were: persistence of finger imprint, body condition, teat length and teat diameter, udder skin temperature, strength of central ligament, age at first calving, gestation length before the investigation, parity group, semester of calving, test day around calving, udder and reproductive veterinary treatments. Only udder oedema severity, persistence of finger imprint, body condition and central ligament were recorded before calving to avoid stressing the late pregnant animals. Cows close to calving are

housed in groups in a free-stall barn. To measure teat length and teat diameter, the cow must be removed from the group and placed in a grooming chute. The farm protocol strictly prohibits stressing, disturbing or driving late pregnant animals. The freshly calved cows were milked in the calving pen for the colostrum period (5-7 days depending on the individual) using a trolley milking machine. During this week the cow is milked only twice a day. After the colostrum period, the cows are moved to the group of cows being in the involution period. The cows of that group are milked three times a day in the milking house. At the end of the involution period (usually 60 days) the cows are moved to the high production group.

The persistence of finger imprint is considered as the degree of skin firmness. On the course of its determination, the index finger is pressed into the oedematous area and a stopwatch is used to measure the time it takes for the udder skin to return to its original state. This time was expressed in seconds. Both middle hindquarters of the cow's udder must be pressed in with the index finger for at least 2 seconds in succession. Evaluation of both sides also serves as a control for physiological (acute) oedema that appears symmetrically. The two values were averaged in the data processing. The fingerprint test was always performed by the same person. The strength of the pressure applied during the test was determined on a kitchen scale which averaged 25 N and corresponded to a pressure of 5 N/cm<sup>2</sup> in our case. In all cases, the fingerprint test was carried out in the most appropriate place, in the milking parlour at the very start of milking, when the milking cups were applied. The main reason for this is that it is compulsory to follow the farm's protocol which does not support the tying of animals in the barn, to expose the cow to unusual situations and to disturb the milking staff. On the other hand, the fingerprint test is simple, can be carried out quickly at the same time as the milking process and can even be carried out by the milking workers later on.

The body condition was defined as the current nutritional status of the individual on the test day. This

was assessed using a scoring system from 1 to 5 (Wildman et al., 1982; Edmonson et al., 1989; DEFRA, 2001).

A tape measure was used to measure the length of the udder teats. The length of the teats is expressed in mm.

The diameter of the udder teats is the width of the nipples at the basis expressed in mm. A digital caliper (MIB 02026065 Digital Caliper 150/0.01 mm DIN 862) was used for measurement. When processing both teat characteristics, data from the front and rear teats were pooled pairwise.

The surface temperature of the udder skin was measured with a non-contact infrared thermometer (model: AOV8711, power: DC3V, accuracy:  $\pm 0.2$  °C, response time: 1 s, measuring distance: 2-5 cm) on the rear udder. The temperature was expressed in Celsius.

The suspension of the udder was determined by the degree of central ligament strength as recommended by WHFF (2005), scored from 1 to 9.

The age at first calving was defined as the age of the individual in days at the time of her first calving.

The length of gestation was defined as the duration of the cow's gestation, which is related to our study, expressed in days.

The parity group in our case gives the total number of calving of cows divided into two classes at the time of the test, where one group includes primiparous (class 1) and the other pluriparous (class 2) individuals.

The semester of calving was determined based on the actual calving date of the individual, dividing the year into two halves according to the experience of large-scale farming in Hungary (code 1 summer half-year: May to October; code 2 winter half-year: November to April; Gáspárdy et al., 2013).

The test days around calving were the days during which cows were visited for evaluation and data recording within the test period around their current calving.

Also, variables considered as potential risk factors for oedema were veterinary treatments related to udder- and reproductive periparturient disorders. Those

treatments based on the criteria of the farm veterinarian were taken into account which occurred within  $\pm 10$  days of the diagnosis of oedema of any severity. Udder- and reproductive biology treatments were taken into account as collective terms. In the case of udder treatment, subclinical mastitis was the most common problem. During reproductive biology treatments, the cows were mostly treated for endometritis, pyometra, retained placenta and vaginal injuries. Even in this grouping, the merging of diagnosed disease forms can be considered valuable.

Production and breeding data (for e.g. calving date, age at calving in days, gestation length) were collected from the Riska farm management system (Systo Ltd., 2014). The complex variables were derived from these.

#### **Statistical analysis**

Taking into account the 62 cows, the basic statistics of the individual fundamental indicators (age at first calving, gestation length, parity group and semester of calving) are given. Subsequently, the basic statistics of the risk factors (including persistence of finger imprint) on oedema each with 294 observations of cows (a cow with an average of 4.7 measurements in each variable) recorded during the whole study period will be prepared. For the latter one-way variance analyses were conducted with oedema score as a grouping variable. Tukey HSD post hoc test was used to detect significant differences among classes of oedema score.

The relationship between each indicator was then assessed by a correlation test, also taking into account each observation.

Then, factor analysis (factor rotation: varimax normalized) was performed to examine the associated variables. The alteration in the risk variable that is a common factor with oedema, as well as the change in the degree of oedema over the study period, is plotted graphically.

From the total population of 62 cows, 50 cows with a maximum udder oedema score of 3 were analysed and compared to the rest of the cows ( $n = 12$ ) with records

of a maximum udder oedema score of 2 during the periparturient period. All variables considered as potential risk factors on the individual highest severity of oedema by use of a logistic regression (logit) with a backward elimination procedure until all variables left in the model had a  $P$  value  $<0.05$ . Parameter estimate, Wald statistics and odds ratio (OR) with 95% confidence intervals (95% CI) were reported.

All the computer processing was conducted using Statistica Version 14.0.0.15 (TIBCO Software Inc., 2020).

## RESULTS AND DISCUSSION

The descriptive statistics of the 62 cows for age at first calving, gestation length, parity group and semester of calving are presented in Table 1. The variables first calving, age and gestation length were set at the lower end of the usual range. The age at first calving in days corresponds to 23.7 months, which characterizes the early maturity of the investigated Holstein-Friesian cows in this herd. The average age at the first calving of the breed in Hungary is 25.01 months (ÁT Ltd., 2021). Hutchison et al. (2017) found that 21-22 months of age is the most ideal age for first calving in Holstein-Friesian heifers. The short gestation period too realizes the need of our days. The distribution of cows in terms of a number of parities was as follows: 48 primiparous cows (77.4%) and 14 multiparous cows (22.6%). The semester of calving showed a more even distribution: 26 cows calved in the summer half of the year (41.8%) and 36 cows calved in the winter half of the year (58.1%).

In Table 2 the basic statistics of the variables recorded during the study period ( $n = 294$  observations) are

collected. The test day shows a confirmed ( $P < 0.001$ ) change in the severity of oedema. Around calving is the most severe cases of oedema observable, and as it moves away from calving the severity of the oedema decreases and eventually disappears.

The body condition score decreases significantly ( $P = 0.029$ ) as the degree of oedema decreases. Studies by Schmidt and Schultz (1959) in cattle and Avais et al. (2020) in dairy goats did not confirm a relationship between udder oedema and body condition. However, this apparent relationship may be due to the energy deficiency typical of the initial part of lactation.

The diameter of the front teats has a proven ( $P = 0.038$ ), positive relationship with the degree of oedema. The length of the front and rear teats showed no association with the degree of oedema. The changes previously observed in acute oedema (Gilbert and Schwark, 1992; Medrano-Galarza et al., 2012; Okkema and Grandin, 2021) were not confirmed in the length of the teats but were confirmed in the diameter of the front teats.

The temperature of the udder skin showed a confirmed ( $P = 0.007$ ), negative relationship with the severity of oedema. In their study, Metzner et al. (2015) observed a decrease in udder surface temperature after treatment of udder quarters infected with *Escherichia coli*, which they attributed to the development of udder oedema. The rise in temperature of the infected udder was observed as a case of pathological oedema by Ramos et al. (2020).

There was found a significant ( $P < 0.001$ ) and positive relationship of the severity of oedema with the persistence of finger imprint.

**Table 1.** Basic statistics of the fundamental variables ( $n = 62$  cows)

Variables	Mean	Median	Minimum	Maximum	sd
Age at first calving, days	723.4	718	666	824	41.9
Gestation length, days	275.6	275	266	284	3.98
Parity group	1.23	1	1	2	0.42
Semester of calving	1.58	2	1	2	0.50

sd – standard deviation

Similar to the findings of Fernandes et al. (2022), there is also no correlation found in this study between udder oedema and udder related treatment (included infections). Bertulat et al. (2012) developed a dynamometer for non-invasive measuring of udder firmness in dairy cows, which could be interesting in mastitis diagnosis or animal welfare control. In their study, Rees et al. (2017) measured udder firmness using this device and found that in cows with mastitis, the lower measurement point of udder firmness was increased, presumably due to udder oedema, a symptom of clinical mastitis.

The literature does not provide data on the relationship between oedema and reproductive-related diseases. According to the basic statistics, it was found that there is a proven ( $P = 0.014$ ), mild positive association between the occurrence of oedema and the frequency of reproductive treatments. Our study shows that oedema as a metabolic

disorder is associated rather with reproductive diseases and treatments. According to Melendez et al. (2006), dystocia is more common in oedematous cows, but there is no difference between oedematous and non-oedematous cows in the other indicator of reproductive performance, the frequency of treatments for metritis.

Table 3 reports the values of the correlation coefficients. The severity of oedema is strongly associated with the test day ( $r = -0.65$ ) and persistence of finger imprint ( $r = 0.66$ ) which is the most important for processing. Waller et al. (2007) reported that oedema was observed for an average of 15 days after calving, with no cows having oedema on day 22 of the study. Helalay et al. (2018) found that the udder turgor of the severely oedematous Holstein-Friesian cows they studied was 15 seconds.

**Table 2.** Basic statistics of the variables recorded during the study period (n = 294 observations)

Variables (P-value)	Oedema score 0 mean $\pm$ sd n = 51	Oedema score 1 mean $\pm$ sd n = 72	Oedema score 2 mean $\pm$ sd n = 83	Oedema score 3 mean $\pm$ sd n = 88
Test day ( $<0.001$ )	30.0 <sup>c</sup> $\pm$ 12.4	16.3 <sup>b</sup> $\pm$ 13.4	8.4 <sup>a</sup> $\pm$ 12.5	3.9 <sup>a</sup> $\pm$ 10.4
Body condition score (0.029)	3.7 <sup>a</sup> $\pm$ 0.71	3.9 <sup>a</sup> $\pm$ 0.75	4.0 <sup>a</sup> $\pm$ 0.76	4.1 <sup>b</sup> $\pm$ 0.74
Front teats length, mm (0.431)	49.2 $\pm$ 9.7	48.9 $\pm$ 10.7	50.9 $\pm$ 10.8	48.2 $\pm$ 10.3
Rear teats length, mm (0.564)	41.9 $\pm$ 6.90	40.8 $\pm$ 9.01	42.1 $\pm$ 7.08	40.6 $\pm$ 7.92
Front teats diameter, mm (0.038)	19.3 <sup>a</sup> $\pm$ 3.37	20.0 <sup>a</sup> $\pm$ 3.66	20.8 <sup>a</sup> $\pm$ 5.78	21.5 <sup>b</sup> $\pm$ 4.71
Rear teats diameter, mm (0.090)	18.5 $\pm$ 3.43	19.1 $\pm$ 3.20	19.8 $\pm$ 4.99	20.2 $\pm$ 4.03
Udder skin temperature, °C (0.007)	36.7 <sup>b</sup> $\pm$ 0.67	36.7 <sup>ab</sup> $\pm$ 0.67	36.6 <sup>a</sup> $\pm$ 0.75	36.4 <sup>a</sup> $\pm$ 0.70
Central ligament score (0.068)	7.0 $\pm$ 1.46	7.0 $\pm$ 1.46	6.9 $\pm$ 1.30	6.5 $\pm$ 1.21
Persistence of finger imprint ( $<0.001$ )	2.5 <sup>a</sup> $\pm$ 9.1	17.1 <sup>b</sup> $\pm$ 22.4	42.5 <sup>c</sup> $\pm$ 32.4	69.8 <sup>d</sup> $\pm$ 36.4
Udder treatment (0.603)	1.1 $\pm$ 0.27	1.2 $\pm$ 0.36	1.1 $\pm$ 0.31	1.1 $\pm$ 0.30
Reproductive treatment (0.014)	1.2 <sup>a</sup> $\pm$ 0.40	1.5 <sup>b</sup> $\pm$ 0.50	1.3 <sup>ab</sup> $\pm$ 0.47	1.4 <sup>ab</sup> $\pm$ 0.48

<sup>a,b,c,d</sup> – different superscript letters show significant differences (Tukey HSD post hoc test  $P < 0.05$ )

sd - standard deviation

**Table 3.** Correlation coefficients (*r*) between the variables under study (coefficients that are significant at the *P* < 0.05 level and reaching 0.25 are in bold)

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	1.00	0.11	-0.09	<b>0.37</b>	0.00	0.03	0.04	-0.07	-0.06	0.12	0.12	-0.24	-0.11	0.17	0.00	-0.17
2		1.00	<b>0.41</b>	<b>-0.26</b>	-0.06	0.00	-0.03	0.13	0.11	-0.05	-0.03	0.11	-0.18	-0.17	0.11	-0.03
3			1.00	<b>-0.29</b>	0.01	-0.14	-0.23	<b>0.31</b>	<b>0.28</b>	0.12	-0.02	<b>0.26</b>	<b>-0.41</b>	-0.16	0.17	-0.06
4				1.00	0.07	-0.01	-0.00	<b>-0.50</b>	<b>-0.39</b>	0.15	0.13	-0.55	0.11	<b>0.29</b>	-0.04	0.01
5					1.00	<b>-0.65</b>	<b>-0.36</b>	-0.06	-0.04	<b>-0.25</b>	-0.23	0.03	0.09	<b>-0.50</b>	-0.16	-0.13
6						1.00	0.19	-0.02	-0.03	0.15	0.14	-0.17	-0.12	<b>0.66</b>	-0.02	0.02
7							1.00	0.05	0.07	0.09	0.10	-0.12	0.16	0.19	-0.04	0.02
8								1.00	<b>0.77</b>	0.11	0.10	<b>0.42</b>	0.00	-0.18	0.05	-0.16
9									1.00	0.03	0.10	<b>0.33</b>	0.03	-0.19	0.03	<b>-0.27</b>
10										1.00	<b>0.77</b>	-0.12	-0.11	0.22	0.14	-0.01
11											1.00	-0.07	0.04	0.17	0.22	-0.05
12												1.00	-0.10	<b>-0.28</b>	0.09	-0.04
13													1.00	-0.06	-0.19	-0.05
14														1.00	-0.03	0.06
15															1.00	0.11
16																1.00

Legend: 1 Age at first calving, 2 Gestation length, 3 Parity group, 4 Semester of calving, 5 Test day, 6 Oedema score, 7 Body condition score, 8 Front teats length, 9 Rear teats length, 10 Front teats diameter, 11 Rear teats diameter, 12 Udder skin temperature, 13 Central ligament score, 14 Persistence of finger imprint, 15 Udder treatment, 16 Reproductive treatment.

A stronger relationship was observed between the length ( $r = 0.77$ ) and diameter ( $r = 0.77$ ) of the front and rear udder teats.

Similarly, Patel et al. (2016) found the same length of anterior and posterior udder teats. A medium-strong relationship was computed between the length of front and rear teats and calving semester ( $r = -0.50$  and  $-0.39$ , respectively), persistence of finger imprint and test day ( $r = -0.50$ ), between the front teat length and udder skin temperature ( $r = 0.42$ ), number of calving and gestation length ( $r = 0.41$ ), and between central ligament and number of calving ( $r = -0.41$ ). At the time of data collection, it was visible that teats were slightly longer during the summer than during the winter, but this isn't supported by any literature references.

Table 4 shows the maximum number of factors that can be distinguished as a result of factor analysis. Overall, the five factors explain almost two-thirds (64.7%) of the total variance in the whole data set. Of this, the share of factor 2 including oedema is close to 15%. In factor 2, oedema is strongly associated with the persistence of finger imprint and test day. Neither udder nor reproductive treatments are included in this factor. This means that the development of oedema is not associated with any disease or disorder that triggers these treatments.

Factor 1 includes the length of the udder teats, plus the semester of the calving and the udder skin temperature. The temperature of the udder surface may be related to the ambient temperature.



**Table 4.** Results of rotated (varimax normalized) factor loadings (loadings written in bold are >0.650)

Variables	Factor 1	Factor 2	Factor 3	Factor 4	Factor 5
Oedema score	-0.070	0.879	0.099	-0.002	0.009
Age at first calving	-0.274	0.044	0.112	0.092	0.699
Gestation length	0.129	0.050	0.694	-0.059	0.102
Parity group	0.271	-0.111	0.769	0.152	-0.071
Semester of calving	-0.755	-0.010	-0.119	0.216	0.297
Test day	-0.095	-0.836	-0.035	-0.141	0.148
Body condition score	0.107	0.484	-0.314	0.080	0.058
Front teats length	0.846	0.030	0.072	0.152	0.145
Rear teats length	0.813	0.011	0.025	0.119	0.275
Front teats diameter	-0.048	0.166	0.082	0.891	0.037
Rear teats diameter	0.010	0.129	-0.049	0.897	0.067
Udder skin temperature	0.655	-0.185	0.074	-0.121	-0.215
Central ligament score	0.103	-0.071	-0.759	-0.072	0.052
Persistence of finger imprint	-0.338	0.751	0.078	0.141	0.041
Udder treatment	0.035	-0.019	0.255	0.400	-0.349
Reproductive treatment	-0.252	0.102	0.056	0.034	-0.707
Explained variance	2.758	2.383	1.880	1.944	1.393
Proportion of total variance	0.172	0.149	0.118	0.122	0.087
Eigenvalues	3.103	2.614	1.769	1.593	1.277

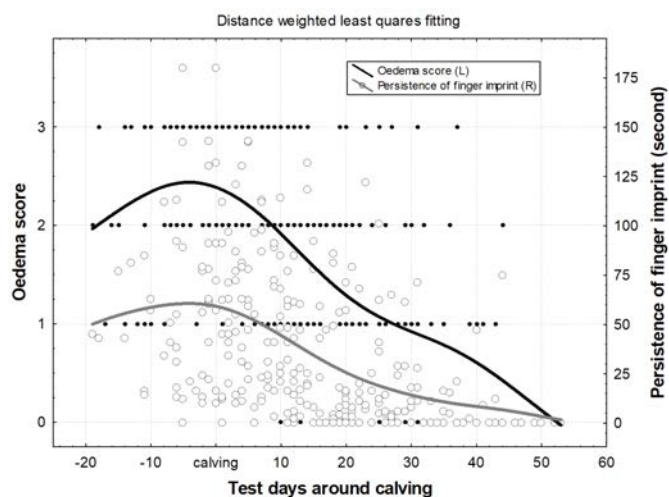
Factor 3 shows the length of gestation, the number of parities and the strength of the udder ligament associated. These variables may be related to the cow's longevity and wear rate. Factor 4 considers only the diameters of the front and rear udder teats to be related. Factor 5 shows reproductive treatment together with the first calving age. These indicators are used to characterize breeding and reproduction, so it is not surprising that it was found a relationship between them.

Figure 1 shows the three variables that are related in factor 2. It can be seen clearly the very similar slopes of the oedema scores and the seconds of finger imprint's persistence according to the test day. As illustrated by the least squares fitting, they reach their maximum value

already a few days before calving. Then, by the 50th day after calving, both fall back to undetectable levels. This confirms that our study included individuals with oedema of physiological origin. Persistent (chronic) oedema can be diagnosed throughout lactation and is usually ventrally localized and is a risk factor for the development of clinical mastitis.

There is no oedema if the skin immediately returns to its original condition and there are no pits. In our experience, for mild oedema (score 1), the slight indentation (about 2-3 mm) disappears around 20-25 seconds. For moderate oedema (score 2), the slightly deeper indentation (about 4-6 mm) persists for about 45-50 seconds. If the oedema is severe (score 3), resulting

in a deep pit (8-10 mm), it is estimated (see Figure 1 and estimate of Table 5) that at least 70-75 seconds on average will be needed for the skin surface to return to its original state.



**Figure 1.** The distribution of oedema scores and seconds of finger imprint's persistence according to the test days

When using logistic regression, of all the risk factors considered, only the persistence of finger imprint showed a significant relationship with the degree of oedema. The Chi-square value (12.2445,  $df = 1$ ) for the difference between the current model (48.68) and the intercept-only model (60.92) was highly significant ( $P < 0.001$ ). Thus, it is concluded that severity of the outcome of oedema is significantly related to the persistence of finger imprint, the firmness of the udder skin. The parameter estimate, Wald statistics and odds ratio (OR) with 95% confidence intervals are presented in Table 5.

**Table 5.** Summary of logistic regression modelling for risk factor skin persistence of finger imprint of the severity of the outcome of udder oedema in oedematous cows

Risk factor	Estimate	Wald P-value	Odds ratio	95% CI OR <sup>1</sup>
Persistence of finger imprint	0.0412	0.0041	1.0421	1.0126-1.0725

<sup>1</sup> lower and upper 95% confidence interval odds ratio.

The parameter estimate can be interpreted as in standard linear regression, that is, a one-second increase in the time it takes for the skin to return to its original

state increases the severity of oedema by 0.0412 points. The odds ratio was 1.0421. An odds ratio that is greater than 1 indicates an improved classification beyond what would be anticipated by random chance. Therefore, the persistence of finger imprint is longer, and the skin is most firm, in the case of the most severe oedema (score 3) in contrast to moderate oedema (score 2). The per cent correct total classification was 87% (while it was 98 for oedema score 3).

## CONCLUSION

The aim of this study is to elaborate a continuous rating method (persistence of finger imprint) to quantitate oedema severity that is fast, efficient, cost-effective, and easily integrated into the daily milking routines at typical Holstein-Friesian large-scale dairy farms in Hungary and supports the visual assessment system of Morrison et al. (2018). A further aim to determine the relationship between the persistence of index finger imprint and the udder oedema scores and the other examined traits (like age at first calving, gestation length, body condition score, front teats length, rear teats length, front teats diameter, rear teats diameter, udder skin temperature, central ligament score and variables (like parity group, semester of calving, test day, udder treatment, reproductive treatment) as well. This study aimed to elaborate a physical examination method (persistence of finger imprint) to quantitate oedema severity for assessing udder oedema, in addition to the visual assessment.

A strong positive relationship was found between the persistence of index finger imprint and the severity of oedema. The more severe the degree of oedema, the more persistent the indentation made by the fingertip. This suggests that the persistence time of the finger imprint can be used to assess the severity of oedema.

A second aim was to determine the relationship between the persistence of finger imprint and the udder oedema scores and the other examined traits and variables.

No correlation was found between oedema and all traits and variables investigated (except persistence of

finger imprint,  $r = 0.66$ ). At the same time, a significant negative association was revealed between the severity of oedema and the test day ( $-0.65$ ), and the persistence of finger imprint and test day ( $r = -0.50$ ).

In the future, to enhance the robustness of findings, it is recommended to undertake further investigations involving additional dairy farms and tests. Furthermore, it is advisable to test the dynamometer developed for measuring udder firmness in cows with mastitis in relation to oedema. A self-developed dynamometer is in progress. By applying constant pressure, it will be possible to create a depression on the skin of the udder. At the same time, in order to ensure repeatability of the measurements, the fixing (base) point of the device is also constant.

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