Effects of a dietary supplement, the MUT, on the urinary parameters in gestating sows

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SUMMARY

This study was carried out on 37 gestating sows to determine the effect of a plant based dietary supplement (MUT) on their urinary status. This product was distributed for 8 days by “top feeding”. Urine samples were taken 6 days before (D-6) and 8 days after the beginning of the supplementation (D7). Five physico-chemical and bacteriological parameters were analysed. On D-6, 29.7% of sows sampled had doubtful results or were infected (bacterial count equal to or greater than 10^4 micro-organisms/ml of urine). After 8 days of receiving MUT, the number of healthy sows had increased by 16.2%. Urinary infection persisted in those sows where the bacterial count was equal to or higher than 10^6 micro-organisms/ml of urine on D-6 (13.5%). Feeding with MUT had normalised certain physical urinary parameters in sows with a normal bacterial count on D7: colour in 65.6% of the sows (p < 0.0001), turbidity for 43.6% of them (p < 0.001) and pH in 31.3% of the animals (p < 0.05). As MUT improves the urinary status of doubtful or mildly infected gestating sows, it may be used as a dietary supplement for comfort in breeding units.

Keywords : sows, urinary infection, dietary supplement, bacterial count, physico-chemical analysis.

RÉSUMÉ

Effets d’un supplément alimentaire, le MUT, sur les paramètres urinaires des truies gestantes.

Cette étude a été réalisée sur 37 truies gestantes afin de déterminer l’effet d’un supplément alimentaire (MUT) à base de plantes sur leur statut urinaire. Ce produit a été distribué durant 8 jours en « top feeding ». Des prélèvements urinaires ont été réalisés 6 jours avant (J-6) et 8 jours après le début de la supplémentation (J7). L’analyse de 5 paramètres physico-chimiques et bactériologiques a été effectuée. A J-6, 29.7% des truies prélevées étaient douteuses ou infectées (numération bactérienne égale ou supérieure à 10^4 germes / ml d’urine). Après 8 jours de distribution du MUT, le nombre de truies saines a augmenté de 16.2%. L’infection urinaire a persisté chez les truies dont la numération bactérienne était égale ou supérieure à 10^6 germes / ml d’urine à J-6 (13.5%). La distribution du MUT a permis de normaliser certains paramètres urinaires physiques chez les truies présentant une numération bactérienne normale à J7 : la couleur pour 65.6% des truies (p < 0.0001), la turbidité pour 43.6% d’entre elles (p < 0.001) ainsi que le pH pour 31.3% des animaux (p < 0.05). Le MUT, en améliorant le statut urinaire des truies gestantes douteuses ou peu infectées, peut être utilisé comme complément alimentaire de confort dans les élevages.

Mots-clés : truies, infection urinaire, supplément alimentaire, numération bactérienne, analyse physico-chimique.

Introduction

In 1983, an inquiry revealed that 20% of French breeding units studied had worrying urinary problems [20]. Another study in 1984 stated that for a total of 3600 sows in around fifty French breeding units, 132 died in a single year [17]. The causes of mortality varied widely but problems concerning the urinary system represented more than 40% of the cases. The cystitis/pyelonephritis complex is one of the main causes of mortality in sows. This is also a breeding unit pathology which may affect up to 25% of sows in a given unit, with different degrees of severity [21]. This proportion is higher than those reported in Great Britain [10], Canada [3] and Denmark [4], where rates vary from 8 to 15.2%.

The usual behaviour of sows is significantly modified when urinary problems appear in an acute form. However, chronic urinary disorders, apparently well tolerated by the animal, upset farrowing and reproductive performance. This supports the usefulness of early screening for urinary disorders [20]. The urine of a sow with no urinary problem has a bacterial count of not more than 10^3 organisms/ml of urine [19, 26], no nitrites [20] and a colour between yellow and amber depending on the concentration of urochromes. Abnormal urine coloration (reddish-brown) may be observed in cases of cystitis/pyelonephritis [8]. Physiological urinary pH varies between 5.5 and 7.5 depending on the animal’s metabolism and the composition of the feed. Significant alkalisation of the urine may indicate an urinary infection [8]. Normal urine is generally clear. It may be cloudy when it contains any of the following : leucocytes, red blood cells, epithelial cells, bacteria, mucus, fat or crystals. A high bacterial load is frequently associated with a positive reaction for nitrites [19]. Identification of urinary disorders is based on a clinical examination including observing vulvar discharges and their microscopic examination and observing miction (the behaviour of the sows and miction volumes). In addition, the physico-chemical parameters of the urine may also be evaluated : colour, turbidity, pH, nitrites and bacterial count [26]. The use of organic acids in animal feed or human food can prevent or treat infection of the urinary tract [1, 24], suppress bacteriuria [30] and decrease urinary pH value [22].

Likewise, several studies show that some botanicals can act in the urinary tract. The effects can be a reduction of urinary infection rates [29], a decrease of urinary pH value [13], a diuretic effect [2, 11, 16] or an antinephrotoxic action [29]. In this study, carried out in a breeding and fattening unit (in France), we tested the effect of a dietary supplement (MUT, a mixture of plants and acidifiers) on five urinary physico-chemical and bacteriological parameters of gestating sows.

**Material and methods**

**ANIMALS AND PROTOCOL DESIGN**

The trial took place in June 2002 in a breeding and fattening unit of 440 crossbreed Large-White-Landrace sows (Poitou-Charente, France). The herd of sows is managed as a fattening unit of 440 crossbreed Large White-Landrace sows under the name MUT (Mix for Urinary Tract). Fifty grams per day were distributed to each sow for 8 days (D0 to D7) by the pig-breeder by "top feeding" on the ration of feed.

Urine samples were taken 6 days before distribution of the MUT (D-6) and 8 days after the beginning of distribution (D7). For urine sampling, the first urine of the day was collected, before distribution of the first meal. The urine was collected in sterile tubes during spontaneous miction, in mid-flow. The samples were taken to the laboratory three hours after sampling and analysed within the day.

**URINE ANALYSIS**

Several urine parameters were analysed:

- **The bacterial count**: was the first step of the analysis, to avoid contamination of the samples during other examinations. It was carried out on Drigalski agar inoculated using a 10 μl calibrated loop. Reading took place after 24 hours of incubation at 37 °C. Counts were determined by comparison with reference standards. The results were expressed as the number of enterobacteria per ml of urine with different values which, in colony forming unity (cfu)/ml, were: < 10^3 ; 10^3 ; 10^4 ; 10^5 ; 10^6 and over. If the count was not more than 10^3 cfu/ml urine, the sow was considered to be healthy [26], while for a value between 10^4 and 10^5 cfu/ml, its state of health was doubtful or little infected and for a value equal or above 10^6 cfu/ml, it was highly infected.

- **For colour determination**, a visual method was used based on a three-levels colorimetric scale: pale clear, mid-yellow, dark yellow. The urine samples were grouped into two classes for this parameter: pale yellow urine and darker yellow urine (mid-yellow + dark yellow).

- **For turbidity**, a visual method was used. The McFarland calibrated standards (BioMérieux) were used to determine 4 degrees of turbidity: clear, slightly cloudy (McFarland 0.5), cloudy (McFarland 1), very cloudy (McFarland 2). The clear urine samples were compared with the cloudy urine samples (grouping together McFarland 0.5 to 2 urine) for analysis of this criterion.

- **The nitrite test**: was performed using dipsticks (Merck Eurolab); the result was either positive or negative.

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat</td>
<td>23.8</td>
</tr>
<tr>
<td>Barley</td>
<td>40.0</td>
</tr>
<tr>
<td>Maize</td>
<td>2.0</td>
</tr>
<tr>
<td>Wheat bran</td>
<td>15.0</td>
</tr>
<tr>
<td>Soybean meal</td>
<td>4.0</td>
</tr>
<tr>
<td>Sunflower meal</td>
<td>7.7</td>
</tr>
<tr>
<td>Cane molasses</td>
<td>2.0</td>
</tr>
<tr>
<td>Citrus pulp</td>
<td>2.6</td>
</tr>
<tr>
<td>Dicalcium phosphate</td>
<td>0.27</td>
</tr>
<tr>
<td>Calcium carbonate</td>
<td>1.41</td>
</tr>
<tr>
<td>NaCl</td>
<td>0.49</td>
</tr>
<tr>
<td>Biolyse 65</td>
<td>0.23</td>
</tr>
<tr>
<td>Vitamins and minerals premix*</td>
<td>0.50</td>
</tr>
</tbody>
</table>

**Table I**: Composition and calculated nutrient content of the feed of the gestating sows (feed manufactured by the firm Peigné, Landemont, France).

A batch of thirty-seven gestating sows was randomly selected from the breeding unit's sows disregarding how many litters they had produced. The sows received the dietary supplement (a mixture of aromatic plants and organic acids) supplied by the company NEAR (Orvault, France) under the name MUT (Mix for Urinary Tract). Fifty grams per day were distributed to each sow for 8 days (D0 to D7) by the pig-breeder by "top feeding" on the ration of feed.

The *pH* was measured using dipsticks. The *pH* values were determined using a colorimetric scale and varied from 5 to 8.5 with intervals of 0.5. For analysing this parameter, a distinction was made between urine samples with pH not more than or equal to 7 and those with a pH greater than 7.

**STATISTICAL ANALYSIS**

The distribution of sows in the study according to bacterial count, urinary turbidity, colour and *pH* between D-6 (before MUT distribution) and D7 (7 days after the beginning of MUT supplementation) were compared using Chi-square tests improved by the Yates correction (the Statgraphics® Plus 5 program was used). This test was also used to study the relationship between bacterial count and turbidity on the one hand and the nitrite test on the other.

**Results**

**CLASSIFICATION OF SOWS RELATIVE TO THE BACTERIAL COUNTS ON D-6 AND D7**

On D-6, 29.7 % of sows sampled were doubtful or infected (Figure 1). After 8 days of the dietary MUT supplementation, the number of highly infected sows decreased by 2.7 % and the number of healthy sows increased by 16.2 % (chi² = 5.71, p = 0.057) : consequently, on D7, 86.5 % of sows had a healthy status. These sows had been not infected (70.3 %), doubtful or little infected (13.5 %) or highly infected (2.7 %) on D-6. After consuming the dietary supplement (MUT), the urinary infection persisted in five sows highly infected (13.5 %) in which the bacterial count was equal to or higher than 10⁶ cfu/ml of urine on D-6.

**RELATIONSHIPS BETWEEN URINE BACTERIAL COUNTS, NITRITE TESTS, TURBIDITY AND PH VALUES BEFORE THE DIETARY MUT SUPPLEMENTATION**

Eleven out of 37 urine samples analysed on D-6 had bacterial count equal or above 10⁶ cfu/ml. The prevalence of the disease (percentage of doubtful or infected sows among all the sows studied) on D-6 was 29.7 %. Table III presents, on D-6, relationships between bacterial counts, nitrite tests, turbidity and *pH* values.

On D-6, bacterial counts and nitrite tests were significantly correlated (p < 0.05) (Table III). Seven out of 37 urine samples tested had a positive nitrite test and the sensibility was 45.5 % : five out of 11 doubtful or infected urine samples were identified by a positive nitrite test. The relationship between the presence of nitrites and urinary infection holds interest because there were only 7.7 % of false positives. The specificity of this urine parameter (92.3 %) was high (Table III). The positive predictive value (probability of urinary infection among urine samples with a positive nitrite test) indicated that 71.4 % of the urine samples with a positive nitrite test were doubtful or infected. The negative predictive value (probability of no urinary infection among urine samples with a negative nitrite test) was 80 %.

Despite the dietary MUT supplementation, 5 sows, highly infected on D-6, retained high bacterial counts on D7. Table IV presents physico-chemical urine parameters in these sows on D-6 and D7. The changes in physico-chemical parameters were studied (Figure 2) for the sows having a normal bacterial count on D7 (86.5 % of the sows sampled). After 8 days of dietary MUT supplementation, the number of sows with abnormal urine colour (mid-yellow or dark yellow) fell by 65.6 %. The difference between D-6 and D7 is significant (chi² = 26.3, p < 0.0001). Turbidity decreased also significantly (chi² = 12.3, p = 0.0005) : between D-6 and D7, the percentage of sows with cloudy urine fell from 53 % to 9.4 %. While 31.3 % of the sows studied had alkaline urine on D-6, after 8 days of consuming MUT, 100 % of the sows had acid urine (chi² = 9.6, p = 0.002). By contrast, the number of urine samples with a positive nitrite test was not changed between D-6 and D7.

The relationship between bacterial counts and urine turbidity on D-6 was significant (p < 0.05) (Table III). Twenty-two out of 37 urine samples analysed were cloudy. The sensitivity of this urine parameter as compared to bacterial counts was high since 90.9 % of the doubtful or infected urine samples were cloudy and 53.8 % of the sows with bacterial counts lower than 10⁶ cfu/ml had clear urine (Table III). The positive predictive value of this parameter was 45.5 %. The negative predictive value was high since 93.3 % of the clear urine samples were healthy.

Bacterial counts and urinary pH were independent parameters on D-6. Eleven out of 37 sows had a urinary pH value above 7, and 69.2 % of the healthy sows had a urinary pH lower or equal to 7. The false negative rate was 72.7 % (sensitivity : 27.3 %). The positive and negative predictive values of urinary pH as compared to urinary infection were 27.3 % and 69.2 % respectively.

**EVIDENCE OF PHYSICO-CHEMICAL URINE PARAMETERS IN SOWS RECEIVING THE DIETARY MUT SUPPLEMENTATION**

The changes in physico-chemical parameters were studied (Figure 2) for the sows having a normal bacterial count on D7 (86.5 % of the sows sampled). After 8 days of dietary MUT supplementation, the number of sows with abnormal urine colour (mid-yellow or dark yellow) fell by 65.6 %. The difference between D-6 and D7 is significant (chi² = 26.3, p < 0.0001). Turbidity decreased also significantly (chi² = 12.3, p = 0.0005) : between D-6 and D7, the percentage of sows with cloudy urine fell from 53 % to 9.4 %. While 31.3 % of the sows studied had alkaline urine on D-6, after 8 days of consuming MUT, 100 % of the sows had acid urine (chi² = 9.6, p = 0.002). By contrast, the number of urine samples with a positive nitrite test was not changed between D-6 and D7.

Despite the dietary MUT supplementation, 5 sows, highly infected on D-6, retained high bacterial counts on D7. Table IV presents physico-chemical urine parameters in these sows on D-6 and D7. On D7, urine colour and urine pH had normal values whereas nitrite test and turbidity results were in accordance with bacterial counts of these sows.
Early identification of urinary infections in gestating sows is of the greatest interest because it allows severe problems to be prevented, particularly during farrowing. The clinical symptoms of urinary infection appear late but are very serious. Besides the observation of miction and vulvar discharges, studying urinary parameters, and particularly bacterial counts, is a reliable and rapid method for diagnosing urinary infections [26]. A bacterial count of less than or equal to $10^3$ micro-organisms/ml is considered as normal in sows’ urine, the micro-organisms counted resulting from contamination during urine sampling [19, 26]. A number of micro-organisms equal to or greater than $10^5$ per ml indicates the presence of a urinary infection in the sow [5, 15, 26].

Of the gestating sows in the breeding and fattening unit, which were randomly selected before studying the effect of the dietary supplement based on plants and acidifiers (MUT), 21.6% had a urinary infection and 8.1% were doubtful cases based on their urine parameters. The situation of the herd as regards urinary disorders on D-6 was not worrying and was similar to that found in average French off-the-ground breeding units [9, 20, 26].

The bacteriological analysis and the nitrite and turbidity tests that we carried out showed that bacterial count is correlated with nitrite content and turbidity. A high bacterial count is in general linked to cloudy urine and positive nitrite tests. The sensitivity of the nitrite test obtained was better than that demonstrated by MADEC and DAVID [19] and SANSOT et al. [26] (45.5% versus 41.7% and 40% respectively). LAMER [15] obtained a higher relationship between bacterial count and nitrites. Their study revealed that the presence of nitrites indicates a microbial infection in 88% of cases against 71.4% in our work. A positive nitrite reaction in the absence of bacteria is rare, as the number of false positives is generally not more than 5%, confirming the very good correlation between the presence of nitrites and urinary infection [15, 19, 26]. The differences seen between the studies can be explained by urinary conditions varying from one study to the other. Indeed, the production of nitrites depends not only of the initial presence of nitrogen compounds in the urine but also of the existence of a flora capable of transforming them (nitrate reductase positive bacteria such as *Escherichia coli* or Streptococci) and of a prolonged retention of the urine in the bladder [9, 18].

The relationship between the bacterial count and urine turbidity is significant and has a sensitivity of 90.9%. These results are better than those reported by LAMER [15] and by GUEROULT [9] (90.9% versus 86% and 89% respectively). The absence of turbidity reflects the absence of a high bacterial count in 93.3% of the cases in our study and in 76% of cases in the study by SANSOT et al. [26].

![Figure 2](image_url)

**Figure 2**: Physico-chemical parameters of urine in sows having “normal” urine bacterial counts (below $10^4$ cfu/ml) the 6th day before (D-6) and the 8th day after (D7) the beginning of the dietary MUT supplementation. Bars with no common superscript differ significantly.

### Discussion

Early identification of urinary infections in gestating sows is of the greatest interest because it allows severe problems to be prevented, particularly during farrowing. The clinical symptoms of urinary infection appear late but are very serious. Besides the observation of miction and vulvar discharges, studying urinary parameters, and particularly bacterial counts, is a reliable and rapid method for diagnosing urinary infections [26]. A bacterial count of less than or equal to $10^3$ micro-organisms/ml is considered as normal in sows’ urine, the micro-organisms counted resulting from contamination during urine sampling [19, 26]. A number of micro-organisms equal to or greater than $10^5$ per ml indicates the presence of a urinary infection in the sow [5, 15, 26]. However, differences are seen in the numbers of bacteria chosen to determine whether or not a sow has a urinary infection. They can be due to the consideration of the degree of pathogenicity specific to each bacterium isolated from the urine [12] and also the dilution factor in potomanic sows [14].
EFFECTS OF A DIETARY SUPPLEMENT, THE MUT, ON THE URINARY PARAMETERS IN GESTATING SOWS

TABLE IV: Physico-chemical parameters of urine in highly infected sows (bacterial counts above or equal to 10⁶ cfu/ml)

<table>
<thead>
<tr>
<th>Nitrite test</th>
<th>Turbidity</th>
<th>Urine colour</th>
<th>pH values</th>
</tr>
</thead>
<tbody>
<tr>
<td>D-6</td>
<td>-</td>
<td>Clear U.</td>
<td>≤7</td>
</tr>
<tr>
<td>D 7</td>
<td>0</td>
<td>Cloudy U.</td>
<td>7 &gt; 7</td>
</tr>
</tbody>
</table>

It has been shown that the pH of urine may vary depending on the composition of a meal or the time when it was ingested. Urine sampled shortly after a meal may have an alkaline pH [23]. In our study, all sampling was carried out after fasting, thus well separated in time from food intake. On the other hand, it has been shown that feed with an electrolyte balance greater than 350 mEq/kg results in respiratory and metabolic alkalosis and in an alkaline pH for the urine [7]. The feed distributed to the gestating sows in the breeding unit studied had an electrolyte equilibrium of about 150 mEq/kg, which could not be the reason for any alkalisation of the urine. As suggested by SERRANO [27], the alkaline urine produced by certain gestating sows used in the study could have been caused by a urinary tract infection. The bacteria that produce urease (in particular the species Staphylococcus, Proteus and Ureaplasma) metabolise the urea into ammonia responsible for the urine alkalisation. A basic pH may thus lead to the suspicion of cystitis or pyelonephritis [26]. During these infections, the urinary pH may reach values of 8 to 9 [6]. Other studies have established that a urinary infection due to Actinobaculum suis may cause significant alkalisation of the urine (pH 8-9) [8] and that sows with cystitis (Eubacterium suis and Streptococcus sp. mainly isolated) have a urine pH of 9 [28].

Our study has shown that giving the supplement based on plants and acidifiers (MUT) over a period of 8 days improves the urinary status of doubtful or little infected gestating sows, and that their bacterial count becomes normal and their physical parameters improved. These results are in accordance with studies showing the efficacy of some botanicals [29] and some organic acids [1, 24, 30] in improving the urinary status in animals or in humans. In addition, no urinary infection appeared in the gestating sows that were healthy before the intake of MUT in the diet, and for 73.1 % of them, MUT improved at least one physical parameter. This product may be used as a dietary supplement for comfort in breeding units. However, in the gestating sows that were highly infected at the start of the study, receiving MUT in the feed for 8 days did not produce any improvement in their microbiological and physico-chemical parameters. To improve the urinary status of highly infected sows therefore, additional work using a more complex dietary supplement combining MUT with other compounds should be undertaken.

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References


