ACCURACY OF A RAPID ELISA TEST KIT TO MEASURE PROGESTERONE IN SOWS

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Introduction
Despite new applications of ultrasonography, blood progesterone is still a reference diagnostic tool for the assessment of reproductive status in many species, including the sow (2, 5). Techniques based on saliva or fecal samples (3), have been investigated, but only serum semi-quantitative ELISA (1) can be implemented at low costs. This study was designed to validate a new ELISA kit, for rapid field determination of sow progesterone.

Materials and methods
Blood samples were collected in the IFIP experimental herd, from 60 females of various physiological status (table 1). Jugular blood was allowed to clot in dry tubes for 12 hours at 4°C before centrifugation and serum was frozen.

Table 1 Characteristics of samples.

<table>
<thead>
<tr>
<th>Progesterone (ng/ml)</th>
<th>n</th>
<th>Mean (sd)</th>
<th>Min</th>
<th>Max</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non pubertal gilts</td>
<td>18</td>
<td>0 (0)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Oestrus (2-3 days post AI)</td>
<td>23</td>
<td>2.7 (2.8)</td>
<td>0.6</td>
<td>12.7</td>
</tr>
<tr>
<td>Pregnancy (4 - 8 weeks)</td>
<td>19</td>
<td>12.0 (3.3)</td>
<td>7.6</td>
<td>19.2</td>
</tr>
</tbody>
</table>

Reference progesterone values were determined by quantitative radioimmunoassay (RIA) in specialized laboratories (INRA, Tours or ENV, Nantes). Semi-quantitative determinations were performed on the same samples, using the Pig-Reprokit® (CEVA Santé Animale, Libourne). This competitive ELISA includes 2 standards : low (2.5 ng/ml) and medium (5 ng/ml). Standards and thawed samples (1 drop) are placed in antibody-coated microwells for incubation with a progesterone conjugate (15 min in dark, at room temperature). Wells are washed before another 15 min incubation with a substrate-chromogen. Subsequent coloration is visually assessed by comparison with standards. Coloration intensity and progesterone concentration are inversely related. Specificity, sensitivity and accuracy were calculated using RIA as reference and a bimodal categorical scale ( ≤ 2.5 ng/ml ; > 2.5 ng/ml ). ELISA were performed simultaneously on the same samples by two independent observers. Agreement was evaluated using Kappa statistic.

Table 2 Accuracy of semi-quantitative ELISA kit

<table>
<thead>
<tr>
<th>Threshold 2.5 ng/ml</th>
<th>RIA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negatives</td>
<td></td>
</tr>
<tr>
<td>Positives</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
</tr>
</tbody>
</table>

Specificity 84 %
Sensitivity 100 %
Global accuracy 92 %

Discussion and Conclusion
The good accuracy of Pig-Reprokit® confirms recent findings on mares with a similar semi-quantitative ELISA (4) or previous work on sows with different kits (1). Low sensitivity is also reported with these rapid tests for intermediate values (4). However, global accuracy is suitable for rapid, on farm, real-time determination of sow cyclicity. This tool may contribute to better management of hormonal treatments. Further investigation is needed to test various possible applications in sow herds.

References

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