

**P-04-20****Reduction of skatole and indole in fat, liver and lean meat by a feeding approach with charcoal (#528)**

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**Introduction**

In the intestine of swine L-tryptophane is metabolised by anaerobic fermentation to skatole (3-methylindole) and indole. The substances are to some extent reabsorbed through the intestinal mucosa. A reduction of these for boar taint liable substances via variations in the feed is possible [1]. If substances, which were metabolised in liver are reabsorbed, they concentrate in liver, kidney and fat [2]. A clear correlation between the concentration of skatole and indole in the intestine and the concentration of these substances in the fat tissue of pigs was demonstrated [3]. With the increase of sexual hormones, especially androstenone, activity of enzymes which are important for skatole metabolism in liver are inhibited. Therefore, skatole breakdown is reduced [4] and boar taint increases.

Charcoal is known for its particularly good adsorption capacity and its use as feed is permitted in the EU feed ordinance (EG Nr. 68/2013). Therefore, the use of charcoal as a feed additive for the adsorption of skatole and indole is obvious for the reduction or avoidance of boar taint. In this approach, a charcoal was fed to boars to adsorb these substances in faeces and it focuses on the comparison of skatole and indole content in liver, fat and meat of control and charcoal feed group.

**Methods**

Fed charcoal consists of oak, beech, spruce and larch, was pyrolysed in a kiln and was characterised in terms of surface energy (dispers = 10.3, polar = 16.5) and specific surface area (app. 500 m<sup>2</sup>/g) before feeding approach. Furthermore, adsorption capacity of charcoal was investigated *in vitro* with faeces as well as in water (Fig. 1). Based on these results, 5 out of 10 hybrid-boar-piglets (mother: German Landrace x German Edelschwein, father: Pietrain) were fed with 2 w/w % charcoal in addition to the normal feed for the last four weeks of living. The rest was fed with control feed. After 15 weeks of living, boars were slaughtered and lean meat from the loin, fat from the back and liver were analysed according to established protocol: skatole and indole were extracted from 10 g meat or 4 g fat or 4 g liver by acetonitrile, MilliQ, a NaCl- + NaSO<sub>4</sub>-mixture and clean-up consisted of freezing the extract at -80 °C for 10 min. An aliquot of the acetonitrile extract was evaporated under a stream of nitrogen at 40 °C to a residual volume of 0.3 – 0.5 ml and taken up in acetonitrile. Solution was membrane filtered, transferred in a vial and analyzed by UPLC-MS/MS (ACQUITY-UPLC-System, Waters;

API4000, AB Sciex). Quantification is done via an external calibration.

For statistical analysis, One Way ANOVA was conducted followed by a Tukey test (p<0.05).

**Results**

Tab. 1 shows mean indole and skatole contents (µg/kg) of lean meat, fat and liver for individual animals and both groups. One significant difference was exposed between control and charcoal group, namely indole content in fat. Significant differences between animals in one group are caused by high contents of some individual (reasons for that are currently unknown). For both groups, skatole is more present than indole in meat *and vice versa* in liver. Interestingly, in charcoal group in lean meat and fat the double amount of skatole in comparison to indole is present, whereas almost double amount of indole in comparison to skatole is present in control group in liver and fat. Due to the young age of pigs at slaughter, these substances are more present in fat and liver than in lean meat since their concentration in fat and liver is faster [2, 3]. By considering these findings and taking previous results from *in vitro* charcoal characterization (Fig. 1) into account, charcoals' good adsorption capacity is obvious and probably caused by the surface energy and specific surface area. Nevertheless, further analyses, e. g. of *in vivo* taken blood and faeces samples, are needed to investigate charcoals' influence in living organism but also on meat quality. To validate the adsorption capacity, further samples of already examined animals are currently analysed and for additional validation, samples were sent to another institute and will be compared.

**Conclusion**

Charcoal, as a sustainable raw material and side stream, has a positive impact on meat quality of boars in terms of the reduction of indole content in fat.

**References**

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**Notes**

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**Adsorption (%) of skatole and indole in faeces and water**

Figure 1: Adsorption (%) of skatole and indole in faeces and water by addition of 0.5, 1, 2, 3, 4 and 5 % of charcoal.

Notes

| Matrix     |        | Lean meat          |         |                    |         | Fat                   |         |                      |         | Liver                 |         |                       |         |
|------------|--------|--------------------|---------|--------------------|---------|-----------------------|---------|----------------------|---------|-----------------------|---------|-----------------------|---------|
| Group      | Animal | Indole             |         | Skatole            |         | Indole                |         | Skatole              |         | Indole                |         | Skatole               |         |
|            |        | Mean               | St.dev. | Mean               | St.dev. | Mean                  | St.dev. | Mean                 | St.dev. | Mean                  | St.dev. | Mean                  | St.dev. |
| Control    | 1      | 1.73 <sup>a</sup>  | 0.60    | 1.90 <sup>a</sup>  | 0.50    | 16.70 <sup>ab</sup>   | 3.15    | 18.20 <sup>a</sup>   | 1.05    | N / A                 | N / A   | N / A                 | N / A   |
|            | 2      | 12.88 <sup>b</sup> | 4.59    | 4.42               | 1.63    | 38.17                 | 5.13    | 27.93 <sup>abc</sup> | 3.50    | 35.13 <sup>a</sup>    | 3.72    | 6.92 <sup>b</sup>     | 1.35    |
|            | 3      | 9.58               | 0.96    | 5.65 <sup>ab</sup> | 1.25    | 38.67                 | 3.16    | 23.80                | 1.59    | 21.67 <sup>ab</sup>   | 3.15    | 17.13 <sup>ab</sup>   | 0.85    |
|            | 4      | 3.17               | 0.69    | 1.57 <sup>b</sup>  | 0.15    | 131.00 <sup>a</sup>   | 26.96   | 13.00 <sup>b</sup>   | 2.19    | 44.33 <sup>b</sup>    | 12.54   | 30.27 <sup>abc</sup>  | 5.76    |
|            | 5      | 2.70               | 0.46    | 3.20               | 0.51    | 18.53 <sup>b</sup>    | 4.07    | 16.07 <sup>c</sup>   | 1.82    | 23.83                 | 5.66    | 17.53 <sup>bc</sup>   | 0.78    |
| summarized |        | 2.80               | 0.94    | 3.35               | 1.79    | 48.61 <sup>a</sup>    | 44.99   | 19.80                | 5.87    | 30.74                 | 11.23   | 17.96                 | 9.02    |
| Char-coal  | 6      | 2.07               | 0.31    | 7.62 <sup>a</sup>  | 0.44    | 12.77 <sup>b</sup>    | 1.85    | 64.67 <sup>a</sup>   | 17.58   | 24.50 <sup>bc</sup>   | 1.25    | 35.77 <sup>a</sup>    | 2.67    |
|            | 7      | 4.07               | 0.41    | 6.47 <sup>b</sup>  | 1.09    | 14.03 <sup>b</sup>    | 1.68    | 24.10                | 0.99    | 22.47 <sup>bc</sup>   | 5.51    | 19.33 <sup>ab</sup>   | 3.06    |
|            | 8      | 2.23               | 0.10    | 5.12 <sup>bc</sup> | 0.58    | 28.63 <sup>abcd</sup> | 8.68    | 28.53                | 3.18    | 66.27 <sup>abc</sup>  | 5.38    | 13.17 <sup>bc</sup>   | 1.80    |
|            | 9      | 2.90               | 0.86    | 2.77 <sup>bc</sup> | 1.04    | 12.80 <sup>c</sup>    | 3.05    | 20.90                | 5.37    | 11.63 <sup>abcd</sup> | 1.25    | 23.87 <sup>abcd</sup> | 3.53    |
|            | 10     | 2.88               | 1.80    | 0.82 <sup>bc</sup> | 0.23    | 14.80 <sup>d</sup>    | 2.25    | 4.85 <sup>a</sup>    | 0.39    | 56.40 <sup>d</sup>    | 6.88    | 12.23 <sup>abcd</sup> | 0.47    |
| summarized |        | 2.83               | 1.03    | 4.56               | 2.64    | 16.61 <sup>a</sup>    | 7.29    | 29.55                | 23.11   | 36.3                  | 22.3    | 20.9                  | 9.1     |

**Indole and skatole content of control and charcoal group in lean meat, fat and liver**

Table 1: Results of indole and skatole content (µg/kg) in lean meat, fat and liver of 10 animals, which were either in control or charcoal group. One Way ANOVA was carried out between mean values of animals from control and charcoal group of one matrix and either indole or skatole. Significant differences were determined with a Tukey test (p<0.05) and are shown with letters. N/A means no results are existing for this sample.

