



CÁTIA LIDIANE ALVES FERNANDES

**FONTES DE FIBRA NA GESTAÇÃO E LACTAÇÃO E SUA
RELAÇÃO COM A MICROBIOTA INTESTINAL E O
DESEMPENHO DE MATRIZES SUÍNAS**

LAVRAS-MG

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Dissertação apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Zootecnia, área de concentração em Produção e Nutrição de Animais Não Ruminantes, para a obtenção do título de

Dr. Prof. Márvio Lobão Teixeira de Abreu
Orientador

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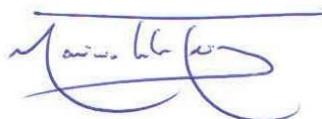
**FONTES DE FIBRA NA GESTAÇÃO E LACTAÇÃO E SUA RELAÇÃO COM
A MICROBIOTA INTESTINAL E O DESEMPENHO DE MATRIZES SUÍNAS**

**SOURCES OF FIBER IN GESTATION AND LACTATION AND THEIR
RELATIONSHIP WITH THE INTESTINAL MICROBIOTA AND THE
PERFORMANCE OF SOWS**

Dissertação apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Zootecnia, área de concentração em Produção e Nutrição de Animais Não Ruminantes, para a obtenção do título de Mestre.

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*Ao meu avô, que estaria orgulhoso em ver aonde eu cheguei e o que o conquistei e a
minha família que sempre me apoiou em todos os momentos*

DEDICO

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“A alegria está na luta, na tentativa, no sofrimento envolvido e não na vitória propriamente dita.”

Mahatma Gandhi

RESUMO GERAL

A gestação é considerada uma das fases mais importante para a produção de suínos, pois seu desempenho pode ser refletido nas fases subsequentes. Estratégias nutricionais como o uso de fibras funcionais podem contribuir para melhorar o desempenho das matrizes suínas durante a gestação e lactação. Objetivou-se avaliar a inclusão de fibras funcionais na dieta e seus impactos na microbiota intestinal, no desempenho das matrizes suínas e de suas respectivas leitegadas, durante a gestação e a lactação. Inicialmente, foram selecionadas 120 matrizes suínas, de 3 a 7 paridades, distribuídas em blocos casualizados. As porcas receberam três dietas durante as fases de gestação e lactação: tratamento controle com inclusão de 15% de casca de soja (CON), tratamento F25S, dieta controle + inclusão de 2,5% na gestação e 1,0% na lactação de fibra parcialmente solúvel e tratamento (FI), dieta controle + inclusão de 2,5% na gestação e 1,0% de fibra insolúvel na lactação. Foram selecionadas 10 porcas de cada tratamento para avaliar as variáveis sanguíneas. Nas 10 porcas também foram avaliados nas fezes os ácidos graxos voláteis (AGV), a microbiota intestinal e o pH, aos 111 dias de gestação e aos 16 dias de lactação. Aos 16 dias de lactação, foram avaliadas as imunoglobulinas IgA e IgG no leite das matrizes suínas. Não houve diferença entre os tratamentos com relação ao desempenho das matrizes suínas e dos leitões ($P>0,05$). Aos 111 dias de gestação, os níveis de ureia plasmática foram superiores no CON comparado com os demais tratamentos ($P<0,10$). A porcentagem de IgG no leite do CON foi superior comparado com F25S e FI ($P<0,05$) e IgA foi maior no F25S ($P<0,05$). Os níveis de ácido acético e isobutírico foram maiores no CON comparados com os demais tratamentos ($P<0,05$). No F25S foi observado maior abundância do filo Euryarchaeota aos 111 dias de gestação ($P<0,10$) e aos 16 dias de lactação ($P<0,05$). Aos 111 dias o gênero *Clostridium* IV e *Methanobrevibacter* foram mais abundantes no F25S ($P<0,10$). No F25S também foi observado menor abundância relativa do gênero *Terrisporobacter* ($P<0,05$). Aos 16 dias de lactação, a abundância relativa dos gêneros *Clostridium* IV e *Clostridium* XIVa foram superiores no CON ($P<0,10$). O gênero *Methanobrevibacter* foi observado em maior abundância no F25S aos 16 dias de lactação ($P<0,10$). Conclui-se que a inclusão de fibras funcionais pode melhorar o aproveitamento dos nutrientes da dieta, a qualidade imunológica do leite, modular a produção de AGVs e alterar a microbiota intestinal das matrizes suínas.

Palavra-chave: Leitegada, Microbioma, Nutrição Funcional, Saúde Intestinal

GENERAL ABSTRACT

The gestation is considered one of the most important stages for swine production, as its performance can be reflected in subsequent stages. Nutritional strategies such as the use of functional fibers can contribute to improve the performance of sows during gestation and lactation. The aim of this study was to evaluate the inclusion of functional fiber in the diet and its impact on the intestinal microbiota, on the performance of sows and their respective litters, during gestation and lactation. Initially, 120 sows were selected, from 3 to 7 farrowing, distributed in random blocks. Sows received three diets during gestation and lactation: control treatment with 15% inclusion of soybean husk (CON), S25F treatment, control diet + 2.5% inclusion during gestation and 1.0% in lactation. partially soluble fiber and treatment (IF), control diet + inclusion of 2.5% during pregnancy and 1.0% insoluble fiber during lactation. 10 sows from each treatment were selected to evaluate blood parameters. In the 10 sows, volatile fatty acids (VFA), intestinal microbiota and pH were also evaluated in feces, at 111 days of gestation and at 16 days of lactation. At 16 days of lactation, immunoglobulins IgA and IgG were evaluated in the milk of sows. There was no significant difference between treatments regarding the performance of sows and piglets ($P > 0.05$). At 111 days of gestation, plasma urea levels were higher in CON compared to other treatments ($P < 0.10$). The percentage of IgG in CON milk was higher compared to S25F and IF and IgA was higher in S25F ($P < 0.05$). The levels of acetic and isobutyric acid were higher in CON compared to the other treatments ($P < 0.05$). In S25F, the highest abundance of the Euryarchaeota phylum was observed at 111 days of gestation ($P < 0.10$) and at 16 days of lactation ($P < 0.05$). At 111 days, the genus *Clostridium IV* and *Methanobrevibacter* were more abundant in S25F ($P < 0.10$). In S25F it was also observed lower relative abundance of the genus *Terrisporobacter* ($P < 0.05$). At 16 days of lactation, the relative abundances of the genus *Clostridium IV* and *Clostridium XIVa* were higher in CON ($P < 0.10$). The genus *Methanobrevibacter* was observed in greater abundance in S25F at 16 days of lactation ($P < 0.10$). It is concluded that the inclusion of functional fibers can improve the use of nutrients present in the diet, the immunological quality of milk, modulate the production of VFAs and alter the intestinal microbiota of the sows.

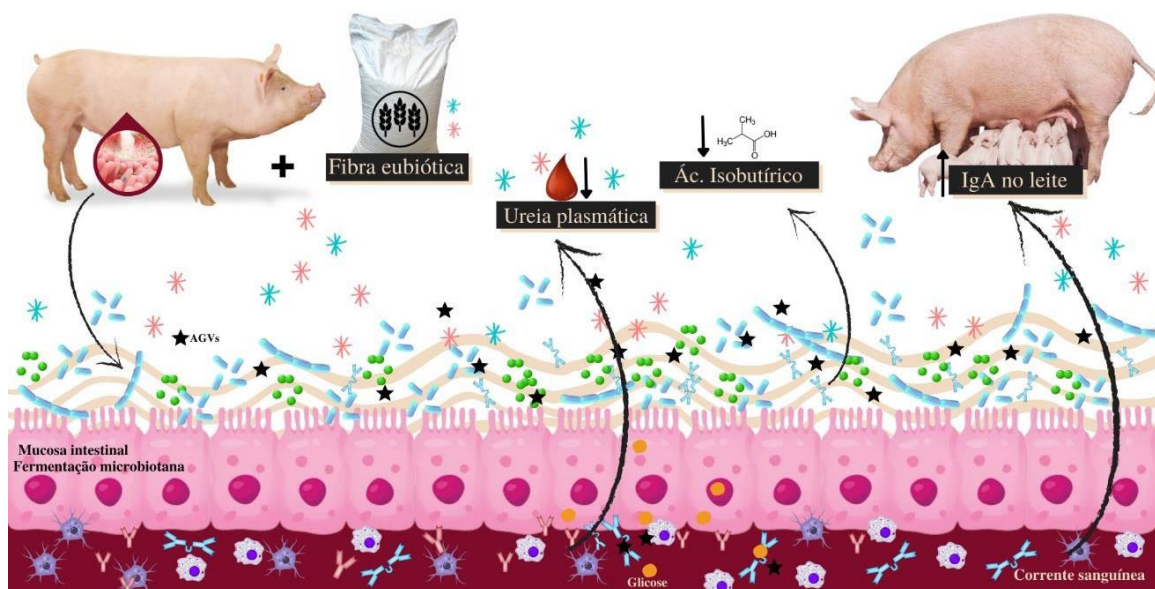
Keywords: Functional Nutrition, Intestinal Health, Litter, Microbiome

Uso de fibras funcionais e sua relação com a microbiota intestinal e o desempenho de matrizes suínas em gestação e lactação

Elaborado por Cátia Lidiane Alves Fernandes e orientado por Márvio Lobão Teixeira de Abreu

O uso das fibras funcionais é uma das estratégias empregadas para melhorar o desempenho das matrizes suínas durante a gestação e lactação. A inclusão de fibras funcionais melhora a absorção dos nutrientes presentes na dieta e auxilia na otimização da motilidade intestinal, minimizando os quadros de constipação. As fibras funcionais têm ação eubiótica, capazes de modular a microbiota intestinal em prol da saúde das matrizes suínas e de suas respectivas leitegadas. Além de modular a microbiota intestinal, as fibras funcionais são capazes de estimular a fermentação microbiana, onde os metabólitos produzidos ativam a resposta imune, por meio do aumento da produção de anticorpos. O estímulo da resposta imunológica materna reflete na imunidade da prole, por meio da transferência de anticorpos no colostro e no leite.

A inclusão de fibra eubiótica foi capaz de alterar a microbiota intestinal das matrizes suínas, estimulando o crescimento de bactérias como as *Methanobrevibacter*, que melhoram a digestibilidade das fibras presentes na dieta e reduz a população de bactérias patogênicas, como as *Terrisporobacter*. As matrizes suínas que receberam dieta com inclusão de fibra eubiótica aumentaram a porcentagem de IgA no leite, garantindo aos leitões maior aporte imunológico.



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PRIMEIRA PARTE

1. INTRODUÇÃO

A suinocultura moderna busca maximizar o desempenho das matrizes suínas, visando o aumento da produção de carne em qualidade e quantidade. E, para o sucesso do sistema de produção é necessário atentar-se à duas fases importantes que o compõem: a gestação e a lactação. Essas duas fases são de grande relevância, uma vez que o desempenho na fase de gestação e lactação das matrizes suínas reflete no desempenho das fêmeas ao longo do ciclo produtivo. A gestação das matrizes suínas tem, em média, 115 dias e é dividida em fases fisiológicas: A implantação embrionária, o crescimento materno e, no terço final de gestação, o intenso crescimento fetal e o desenvolvimento da glândula mamária.

O periparto caracteriza-se como uma fase delicada para as matrizes suínas. Compreende-se como periparto o intervalo de transição da gestação para a lactação, entre 110 dias de gestação até o décimo dia pós-parto. Nesse período, as matrizes suínas passam por processos metabólicos e fisiológicos que demandam maior aporte nutricional para a finalização da formação fetal para a síntese de colostro e de leite. Além disso, durante o período lactacional as fêmeas suínas passam por intenso catabolismo, resultando em distúrbios metabólicos prejudiciais à saúde (ZHOU et al., 2017). Quando as fêmeas suínas chegam ao periparto com distúrbios metabólicos em decorrência da má condição corporal ou por falhas nutricionais, há uma sobrecarga metabólica nesta fase, o qual pode comprometer o desempenho lactacional (LI et al., 2019).

O uso de alimentos funcionais tem sido constantemente estudado, principalmente a inclusão de fibras como estratégia nutricional, com o intuito de otimizar os índices zootécnicos e econômicos na produção animal. Uma das estratégias empregadas durante a fase gestacional e lactacional é o uso de fibras funcionais. A inclusão de fibras funcionais melhora a digestibilidade dos nutrientes presentes na dieta e auxilia na otimização da motilidade intestinal, minimizando os quadros de constipação. Além disso, as fibras funcionais têm ação eubiótica ou estímbiótica, capazes de modular a microbiota intestinal em prol da saúde das matrizes suínas e de suas respectivas leitegadas (SHANG et al., 2021).

Uma microbiota intestinal em eubiose está associada à melhor aproveitamento dos nutrientes, bem como a produção de metabólitos que atuam na saúde intestinal. Além de

modular a microbiota intestinal, as fibras funcionais são capazes de estimular a fermentação microbiana, onde os metabólitos produzidos ativam a resposta imune, por meio do aumento da produção de anticorpos. O estímulo da resposta imunológica materna reflete na imunidade da prole, por meio da transferência de anticorpos no colostro e no leite (KIM et al., 2016).

A inclusão de fibras na dieta pode ser empregada como estratégia nutricional que melhora o status metabólico e a saúde intestinal das matrizes suínas, principalmente em uma fase tão estressante como o periparto. Portanto, essa pesquisa foi desenvolvida com o intuito de avaliar os efeitos da inclusão de fibras funcionais nas dietas nas fases de gestação e lactação e a relação com a microbiota intestinal e com o desempenho lactacional das matrizes suínas.

2. REFERENCIAL TEÓRICO

2.1 Gestação de matrizes suínas

O período de gestação é considerado uma das principais fases do sistema de produção de suínos. O desempenho das matrizes suínas durante a gestação reflete nas fases subsequentes do sistema de produção (TROTTIER et al., 2015). Basicamente, a gestação é dividida em fases: implantação embrionária e estabilização da gestação, até 28 dias após a cobertura, crescimento materno e recuperação das reservas corporais, do 29º dia até os 85 dias de gestação, e intenso crescimento fetal e desenvolvimento da glândula mamária, dos 86 dias de gestação até o parto (LANGENDIJK, 2015; SOLÀ-ORIOU e GASA, 2017).

A implantação embrionária é uma fase delicada e decisiva, onde é definido o tamanho da leitegada, pelo número de embriões que sobreviveram. No terço médio, é a fase em que as fêmeas podem se recuperar dos desgastes do parto anterior ou de severas perdas corporais, por meio do controle alimentar. No terço médio também ocorre a miogênese fetal, processo fisiológico essencial para o desempenho dos leitões ao longo das fases de produção subsequentes (PARDO et al., 2013).

O periparto caracteriza-se como um período delicado para a saúde das matrizes suínas, em decorrência dos inúmeros fenômenos que ocorrem nesta fase (FEYERA e THEIL, 2017). Nessa etapa, as porcas passam por alterações metabólicas e fisiológicas

severas que demandam elevado aporte nutricional (OLIVIERO et al., 2010). De acordo com Anil et al. (2006), o periparto é o período de maior risco do ciclo reprodutivo, com 42% das mortes de porcas ocorrendo durante esse curto intervalo.

Outro evento ocorrido no periparto é o acelerado desenvolvimento da glândula mamária. No momento em que antecedente ao parto, Ji et al. (2006) relataram que há um aumento no peso da glândula mamária até os 112 dias de gestação, frente à necessidade da produção de colostro. Entretanto, o desenvolvimento da glândula mamária não para no final da gestação, estendendo-se até a lactação, com a proliferação celular dos lactócitos, células responsáveis pela produção de leite (FARMER e HURLEY, 2015).

A produção do colostro tem início antes do parto, porém não se sabe exatamente quando isso ocorre. Atualmente, acredita-se que boa parte da síntese de colostro aconteça nos últimos 10 dias de gestação (THEIL et al., 2014). Alinhado a isso, alguns trabalhos relataram o aumento da concentração de lactose no plasma sanguíneo da porca nos últimos quatro dias antes do parto, indicando que a lactose colostrada é produzida nos últimos dias da gestação (HARTMANN et al., 1984; HURLEY, 2003). Outro marcador encontrado no plasma de porcas no pré-parto foi a lactoalbumina, a qual participa do complexo enzimático que catalisa a síntese de lactose (DODD et al., 1994).

O momento do parto é considerado o processo fisiológico mais estressante para a fêmea, e a sua duração pode ser crucial para a saúde da porca e de sua respectiva leitegada (OLIVIERO et al., 2019). Leitegadas maiores prolongam o tempo de duração do parto, afetando negativamente a saúde da porca e dos leitões recém-nascidos (BJÖRKMAN et al., 2017). Outro fator que dificulta o processo do parto é a condição corporal da porca, sendo porcas gordas mais propícias à partos distócicos e à redução no número de leitões nascidos vivos (LAVERY et al., 2019; OLIVIERO et al., 2010; ROONGSITTHICHAJ e TUMMARUK, 2014).

Sendo assim, existem programas nutricionais, alinhados a uma ótima condição corporal no momento do parto, que contribuem para a produção de leitegadas mais pesadas e leitões com maior peso ao nascimento, além de atenderem a demandas metabólicas para a produção de colostro e de leite (DECALUWE et al., 2013; CERUTTI e VERARDI, 2014).

2.2 Nutrição das matrizes suínas na gestação

O progresso da nutrição das matrizes suínas deve-se principalmente pela necessidade de adequar os programas nutricionais ao potencial genético, e ao nível de produção das fêmeas disponíveis atualmente mercado (GAILLARD et al., 2019). Essas fêmeas são mais precoces, mais produtivas e mais exigentes nutricionalmente (EISSEN et al., 2000; TROTTIER et al., 2015; BIDANEL et al., 1994; TOWN et al., 2004). Além disso, possuem menor reserva corporal de gordura, o que por consequência apresentam maior perda da condição corporal, resultando na redução da produtividade e em falhas reprodutivas (KIM et al., 2015b). A nutrição das matrizes suínas deve estar devidamente relacionada à ordem de parto, à genética, à condição corporal e aos fenômenos metabólicos e fisiológicos ocorridos ao longo da gestação (THEKKOOT et al., 2016; SEOANE et al., 2020).

As alterações na demanda nutricional das porcas estão diretamente ligadas às modificações metabólicas, em resposta à deposição proteica e lipídica como reserva tecidual nos diferentes estádios da gestação (THOMAS et al., 2018; SEOANE et al., 2020). O aumento da exigência de aminoácidos no final da gestação pode ser explicado devido ao intenso desenvolvimento fetal e da glândula mamária nesse período (SEOANE et al., 2020). De acordo com os estudos conduzidos por Samuel et al. (2012), há um aumento substancial na exigência de lisina até o terço final da gestação, o que enfatiza a importância da alimentação em fases para atender as exigências nutricionais das fêmeas gestantes.

À medida que a gestação progride, há um aumento na demanda energética das porcas (NRC, 2012). Logo, a ingestão insuficiente de energia pode provocar a mobilização lipídica das reservas corporais para a utilização como fonte energética, principalmente no final da gestação (DOURMAD et al., 1996). Entretanto, estudos realizados por Gonçalves et al. (2016) relataram maior porcentagem de leitões natimortos em leitegadas de porcas que receberam elevados níveis de energia na dieta.

Na gestação, adota-se a restrição alimentar como medida de controle da condição corporal, limitando o excesso de reservas corporais, o qual garante melhores resultados futuros no parto e na lactação (HUBER et al., 2019). Vignola (2009) relatou em seu estudo que fêmeas suínas superalimentadas, principalmente nas primeiras duas semanas após a inseminação, apresentaram alta mortalidade embrionária e produziram leitegadas

menores. Já nas últimas semanas de gestação, a superalimentação pode ter influência positiva no peso ao nascimento dos leitões, devido à elevada concentração de nutrientes disponíveis para os fetos (VELA BELLO et al., 2015). O consumo de ração no terço final da gestação deve ser ajustado nutricionalmente para minimizar o balanço energético negativo ocorrido no periparto, e promover maior ingestão de ração no início da lactação (VIGNOLA, 2009).

No terço final da gestação, o rápido crescimento da prole, o desenvolvimento mamário e o parto, são alguns dos fatores que provocam estresse metabólico, dando início ao quadro de balanço energético negativo, forçando a mobilização de reservas proteicas e energéticas do organismo (REMPEL et al., 2015; THEIL et al., 2014). Por consequência, a demanda nutricional não é atendida para sustentar os processos fisiológicos ocorridos no periparto, o que gera impactos na produtividade das matrizes e de suas respectivas leitegadas nas fases subsequentes do sistema de produção (THEIL, 2015; QUESNEL et al., 2012).

O manejo nutricional na gestação tem papel crucial para a saúde e produtividade das matrizes suínas, principalmente durante o periparto (PRUNIER et al., 2010). Sendo assim, vale salientar a importância dessa etapa para o sistema de produção, visto que, esses fenômenos podem acarretar perdas no desempenho lactacional das matrizes suínas (TOKACH et al., 2019).

2.3 Nutrição na gestação e seu impacto na lactação

Sabe-se que os cuidados nutricionais das matrizes suínas não devem se limitar apenas na gestação, mas sim ao longo da vida produtiva, respeitando a exigência nutricional de cada fase (SOLÀ-ORIOLE e GASA, 2017). O consumo de ração no terço final da gestação deve ser ajustado nutricionalmente para minimizar o balanço energético negativo ocorrido no periparto e promover maior ingestão de ração no início da lactação (VIGNOLA, 2009). No início da lactação, o consumo de ração não é suficiente para atender as demandas corporais, fazendo com que haja a mobilização corporal para atender o metabolismo basal da produção de leite das matrizes suínas (MOSNIER et al., 2010; HANSEN, 2012a).

Na fase de transição entre a gestação e a lactação, aproximadamente aos 110 dias de gestação, as matrizes suínas devem ser transferidas para o galpão de maternidade, onde

receberão ração de lactação ou de transição à vontade até o momento do parto (SILVEIRA e ZANELLA, 2014). Nessa fase, é necessário que a ingestão de ração seja adequada para sustentar a demanda cada vez maior dos fetos e das porcas, melhorando a captação placentária de nutrientes e oxigênio (BELKACEMI et al., 2010). Neste caso, as dietas de transição consistem em rações de pré e pós-parto, com maiores níveis de energia e nutrientes que melhoram a saúde das porcas no momento do parto (FEYERA et al., 2018). Essas dietas contribuem para a ocorrência de partos mais rápidos, aumento no número de leitões nascidos vivos, maior vitalidade da leitegada e conseqüentemente, aumento no número de desmamados (FEYERA e THEIL, 2017; THEIL et al., 2014;).

A alimentação pode desempenhar um papel importante, principalmente no momento do parto (PEDERSEN, 2019). Uma das estratégias nutricionais é a inclusão de fibra na dieta pré-parto para aliviar o desconforto causado pela constipação nos dias próximos ao parto (JIANG et al., 2017). Este desconforto é resultante da redução no consumo de água e o aumento da absorção hídrica do organismo no momento próximo ao parto (JANG et al., 2015; SONG et al., 2015).

2.4 Fibras na alimentação de suínos

A fibra pode ser denominada como um conjunto de carboidratos estruturais que compõem a parede celular das plantas. Destacam-se os polissacarídeos não amiláceos (PNAs), pectinas, celulose, hemiceluloses, β -glucanos, frutanos e os compostos fenólicos como a lignina, por exemplo. É considerada como a parte do alimento não digerido por enzimas produzidas pelo trato digestório de animais não-ruminantes, porém hidrolisada no intestino grosso por meio da fermentação microbiana (SAKOMURA, 2014).

A fibra pode ser determinada de acordo com a solubilidade dos compostos que a compõe, normalmente utilizando metodologias que separam a fibra em fibra bruta (FB), fibra em detergente neutro (FDN) e fibra em detergente ácido (FDA). Essas metodologias acabam subestimando o valor da fibra na dieta quando mensuram apenas o PNA insolúvel e a lignina, desconsiderando o real teor de fibra solúvel presente na dieta (BACH KNUDSEN, 2001).

As propriedades fisiológicas da fibra também podem estar relacionadas com a viscosidade, estrutura física e capacidade de retenção de água. Classicamente, as fibras dietéticas foram categorizadas como fibras solúveis e insolúveis. As fibras solúveis são fermentadas no cólon para produzir gases e subprodutos fisiologicamente ativos (MAKKI

et al., 2018). As fibras insolúveis são metabolicamente inertes e fornecem volume para a dieta (CAPUANO, 2017). Acredita-se que as fibras solúveis são fermentadas mais rapidamente, produzindo maiores quantidades de ácidos graxos de cadeia curta (AGCC) do que as fibras insolúveis, favorecendo a proliferação da microbiota benéfica (JHA e BERROCOSO, 2015).

Os não-ruminantes em geral, são limitados a digestão das fibras devido à ausência de celulasas no trato digestório (MOLIST et al., 2009). No entanto, a fibra tem importância no processo físico da digestão, influenciando na formação e condução do bolo alimentar e na modulação da microbiota intestinal (TIAN et al., 2020). Além disso, a fermentação microbiana da fibra tem como produto final os gases hidrogênio, metano e dióxido de carbono, e a produção de ácidos orgânicos como AGCC e ácido lático (LI et al., 2020a; TIAN et al., 2020). Estes produtos da fermentação microbiana estão relacionados com a saúde e vias metabólicas do organismo (WILLIAMS et al., 2017).

A utilização da fibra também contribui para alterações fisiológicas e metabólicas, promovendo melhorias na performance reprodutiva das matrizes suínas (CHENG et al., 2015). Os ácidos graxos voláteis (AGVs) oriundos da fermentação da fibra podem controlar a liberação de adipocinas no epitélio intestinal de porcas em crescimento (KIM et al., 2010c). Em humanos e camundongos, os AGVs podem regular a expressão gênica de fatores relacionados a deposição de gordura, porém esse fato não foi bem elucidado em suínos (WEBER et al., 2012). Entretanto, estudos que avaliaram os efeitos da fibra no metabolismo energético de porcas, observaram o controle no depósito de gordura graças ao aumento da sensibilidade à insulina durante a gestação (TAN et al., 2016; ZHOU et al., 2017).

No parto, a fibra pode contribuir para o aumento da absorção de nutrientes e de energia do trato gastrointestinal através da fermentação microbiana (JHA e BERROCOSO, 2016). A fermentação da fibra produz energia disponível para absorção no trato gastrointestinal, regulando os níveis glicêmicos no sangue durante o parto, quando este processo fisiológico exige uma considerável demanda energética (FEYERA et al., 2018).

Além dos efeitos maternos, a inclusão de fibra na dieta de transição das porcas (aos 95 dias de gestação até cinco dias pós-parto) pode aumentar a taxa de crescimento dos leitões nos primeiros dias após o parto, comparado com porcas que não receberam

fibra na dieta (OLIVIERO et al., 2009; QUESNEL et al., 2009). Um dos motivos se deve pelo fato das porcas, quando saciadas pela inclusão de fibra na dieta, permanecerem deitadas por mais tempo, favorecendo o acesso dos leitões ao colostro e leite (OELKE et al., 2018).

A fibra também pode proporcionar o estado de saciedade para as fêmeas, reduzindo movimentos nas gaiolas não considerados como estereotípias (ROBERT et al., 1997). A redução da movimentação das porcas durante a lactação contribui para o aumento na taxa de sobrevivência dos leitões, refletindo nos índices pré-desmames como a porcentagem de leitões desmamados e a queda na taxa de mortalidade (OELKE et al., 2018).

A escolha do programa nutricional adequado na gestação pode maximizar a produtividade das matrizes suínas lactantes, bem como favorecer a produção de leite nessa fase, promovendo maior aporte nutricional aos leitões (FEYERA e THEIL, 2017). Como alternativa, a fibra pode trazer benefícios para o sistema de produção, seja este alimento utilizado como ingrediente ou como alimento funcional (ZHOU et al., 2017; SHANG et al., 2021).

O efeito da fibra no desempenho das matrizes pode variar de acordo com as propriedades físico-químicas distintas de acordo com a fibra utilizada nas dietas. Estes resultados foram encontrados por Weng (2019), onde foram comparados diferentes ingredientes fibrosos durante o período gestacional de fêmeas suínas. Ao avaliar a inclusão de diferentes fontes de fibra, Shang et al. (2019) observaram diferença significativa no consumo de ração de porcas lactantes alimentadas com polpa de beterraba, comparado com os demais tratamentos. O aumento do consumo de ração desencadeou a melhora no desempenho das matrizes e a saúde intestinal dos leitões.

As vantagens do uso de fibra na alimentação animal estão relacionadas além do valor nutricional, à melhor digestibilidade do produto final. Isso ocorre devido à ação das enzimas exógenas produzidas na degradação destes materiais fibrosos, capazes então de promover a hidrólise da glicose e da celulose (celulases), da hemicelulose (xilanase) e da lignina (lacases e peroxidases) (SOUZA, 2012). Urriola et al. (2013) observaram que altos teores de fibra na dieta de suínos diminuíram a digestibilidade proteica, visto que a fibra pode interferir no acesso dos microrganismos intestinais ao conteúdo proteico. Dessa

forma, existe relação entre a digestibilidade dos nutrientes da dieta com o teor de fibra da ração.

Machado et al. (2007) encontraram resultados satisfatórios com a inclusão de fibra, sendo utilizado como um alimento funcional na dieta de frangos de corte para a diminuição do uso de antibióticos. No estudo realizado por Shang et al. (2019), foram encontrados níveis elevados de imunoglobulina A (IgA) e interleucina 10 (IL-10) no colostro de porcas lactantes que receberam inclusão de fibra na dieta, ou seja, maior transferência de componentes imunológicos aos leitões. Além disso, foi observado a expressão de genes responsáveis pela integridade intestinal dos leitões, levando à uma possível redução da necessidade de antimicrobianos contra enfermidades intestinais (SHANG et al., 2019).

Tanto na gestação quanto na lactação, as fêmeas hiperprolíficas entram em catabolismo intenso devido ao alto requerimento por nutrientes que por sua vez, afeta o sistema imune e as habilidades reprodutivas futuras (PRUNIER et al., 2010). Em vista disso, estudos realizados por Xu et al. (2020) sugerem que a ingestão de fibra durante a gestação e lactação melhora a saúde intestinal, atuando na modulação da microbiota intestinal e na resposta imune das fêmeas e de suas respectivas leitegadas.

2.5 Importância da microbiota intestinal

No geral, um intestino saudável é habitado por milhares de espécies de microrganismos que juntos formam a microbiota intestinal (FOUHSE et al., 2016). A saúde intestinal está diretamente ligada ao perfeito funcionamento das barreiras físicas e químicas que compõem o trato gastrointestinal para impedir a invasão de microrganismos e substâncias indesejáveis no epitélio (FAN et al., 2019).

A superfície da mucosa intestinal é protegida por uma camada de mucinas e lipídeos que impedem a exposição das células epiteliais do trato gastrointestinal contra injúrias físicas ou o contato com microrganismos patogênicos (SCHNEIDER et al., 2018). Além da camada que reveste a mucosa, o pH gástrico e as secreções pancreáticas e biliares de enzimas digestivas contribuem para a proteção química contra microrganismos patogênicos e antígenos (VIGGIANO et al., 2015).

O epitélio intestinal também possui estruturas complexas e dinâmicas conhecidas como junções firmes, que regulam a passagem paracelular de íons, solutos e água

(PETERSON, 2014). Essas estruturas compostas por proteínas integrais de membrana exercem função de barreira semipermeável, bloqueando a passagem de proteínas, lipídeos e produtos patogênicos para a corrente sanguínea (FAN et al., 2019). As junções firmes são constituídas pelos grupos de proteínas denominados como claudinas, ocludinas, moléculas de adesão juncional e *zonula occluden* (ZO). Essas proteínas se diferem de acordo com a quantidade de alças que transpassam a membrana plasmática das células (ODENWALD e TURNER, 2017).

O microbioma presente no TGI, quando estável e equilibrado, confere proteção ao hospedeiro, garantindo a integridade das barreiras intestinais, além de contribuir para maior absorção dos nutrientes e na modulação do sistema imune (CLEMENTE et al., 2012). Entretanto, em estado de disbiose, ou seja, quando ocorre alterações na composição e diversidade das bactérias, a microbiota intestinal pode modificar o mecanismo imune intestinal, favorecendo a proliferação e a translocação de bactérias patogênicas para a corrente sanguínea (PETERSON, 2014).

O desequilíbrio da comunidade microbiana intestinal pode estar associado ao desenvolvimento de várias condições, desde inflamações basais e crônicas, doenças intestinais até síndromes metabólicas (BROWN et al., 2012a). Frente à sua importância, estudos têm sido desenvolvidos a fim de encontrar correlações da microbiota intestinal com alguns fatores que agravam a saúde nos animais e que afetam a lucratividade do sistema de produção (MORAN et al., 2014; HASAN et al., 2018; JANSSEN et al., 2015).

Nos últimos anos, o microbioma intestinal do suíno tornou-se o foco de alguns estudos, graças a sua importância para a saúde do hospedeiro (VIGORS et al., 2016; METZLER-ZEBELI et al., 2015). A microbiota intestinal ao longo do trato gastrointestinal, desempenha várias funções simbióticas e mutualistas, não apenas entre os microrganismos, mas principalmente com o hospedeiro, estimulando o sistema imunológico, auxiliando na digestão de alimento e fornecendo substratos importantes, como os AGCCs (STANLEY, HUGHES e MOORE, 2014).

Alguns estudos relacionados à obesidade foram desenvolvidos com o intuito de investigar quais os impactos do sobrepeso na microbiota intestinal e suas alterações no perfil metabólico de humanos e ratos (BOULANGE et al., 2016; ALUTHGE et al., 2019). Entretanto, Xiao et al. (2016) identificaram maior semelhança entre o microbioma de humanos e suínos do que entre humanos e camundongos. Estes autores relataram que

aproximadamente 96% das vias funcionais descritas no microbioma intestinal de humanos também são comuns ao microbioma de suínos.

Além disso, os seres humanos compartilham mais semelhanças com suínos em termos de anatomia, genética, fisiologia, biodisponibilidade farmacêutica e digestibilidade dos nutrientes comparado aos roedores (WANG et al., 2016b). Portanto, o suíno é um modelo animal superior aos roedores para o estudo da fisiologia e patologia humana, no que diz respeito à saúde intestinal.

A formação da microbiota intestinal dos suínos começa no momento do nascimento e pode estar diretamente relacionada com a condição corporal, composição da dieta, processos fisiológicos, *status* metabólico, com o ambiente, entre outros fatores (CHENG et al., 2019; ALUTHGE et al., 2019; HE et al., 2019). Ao longo do TGI, os microrganismos distribuem-se de acordo com as características específicas de cada segmento, com diferenças no pH, e pela presença de enzimas digestivas e de sais biliares que podem alterar a comunidade microbiana de cada segmento intestinal (MACKIE et al., 1999).

Segundo Niu et al. (2019), as porcas apresentam população microbiana intestinal única, comparada com os animais das outras fases do sistema de produção. Estudos realizados por Liu et al. (2019), utilizando fêmeas suínas gestantes como modelo animal, relataram que a abundância das bactérias pertencentes à família Coriobacteriaceae e do gênero *Escherichia* aumentaram gradualmente ao longo da gestação. De acordo com os estudos de Koren et al. (2012), as modificações na composição da microbiota intestinal podem ser influenciadas pela atividade metabólica em mulheres ao longo da gestação. Em alguns trabalhos, a família Coriobacteriaceae tem sido associada com o aumento nos níveis séricos de insulina, c-peptídeo, colesterol total e HDL ao longo da gestação, servindo como biomarcador relacionado ao metabolismo energético prejudicado (MARTÍNEZ et al., 2014; BARTELS e O'DONOGHUE., 2011).

Sendo assim, para comprovar a importância da microbiota intestinal materna, Leblois et al. (2017) realizaram um estudo sobre a composição microbiana do cordão umbilical de leitões e encontraram que a transferência microbiana materna é possível e que pode ocorrer durante a gestação. Além disso, a nutrição materna pode influenciar na concentração de AGVs da digesta da prole. Estes mesmos autores também encontraram maior concentração de AGVs na digesta de leitões nascidos de porcas alimentadas com

dieta rica em fibra, em comparação com porcas que receberam baixa inclusão de fibras na dieta.

Os AGVs são os principais produtos da fermentação bacteriana do trato gastrointestinal (VAL-LAILLET et al., 2017). As enzimas microbianas podem transformar polissacarídeos em fontes de energia digeríveis, particularmente monossacarídeos e ácidos graxos de cadeia curta (KOMAROFF et al., 2017).

Os filos Firmicutes, Bacteroidetes e Proteobacteria são os principais constituintes que compõem a microbiota intestinal de suínos (RAMAYO-CALDAS, 2016; XIAO et al., 2016; ZHAO et al., 2015). As bactérias do filo Firmicutes são conhecidas por produzirem butirato, enquanto as bactérias do filo Bacteroidetes produzem acetato e propionato como produtos do metabolismo primário (MACFARLANE et al., 2003).

O ácido butírico pode atuar inibindo a colonização e proliferação de microrganismos patogênicos no trato gastrointestinal, melhorando a mucosa intestinal, e conseqüentemente, a absorção de nutrientes (HUANG et al., 2015; VIEIRA et al., 2020). A maior eficiência de absorção dos nutrientes aumenta a quantidade de energia canalizada para processos metabólicos, como a produção de leite, reduzindo a mobilização das reservas corporais (THINGNES et al. e BRAHIM et al., 2012).

O gênero *Lactobacillus* é capaz de sintetizar ácido lático e degradar a lactose e outros oligossacarídeos em acetato e lactato (WALTER, 2008). A ordem Clostridiales pode digerir proteínas, carboidratos, açúcar, aminoácidos, purina, pirimidina e outros compostos orgânicos, para sintetizar AGVs (ZHANG et al., 2016). As bactérias das famílias Clostridiaceae, Lachnospiraceae, Ruminococcaceae e as bactérias do filo Prevotella sintetizam boa parte das enzimas degradadoras de carboidrato, resultando na elevada produção de AGCCs (VAN DEN ABBEELE et al., 2013). *Bacteroides* é um gênero caracterizado pela elevada expressão de xilose isomerase. Além de *Bacteroides*, *Parabacteroides* sintetizam ácido propiônico como produto final (MACFABE et al., 2007).

2.6 Fibra funcional e microbiota intestinal

Como estratégia nutricional, a inclusão de alimentos funcionais pode contribuir para a maximização do desempenho produtivo e reprodutivo das matrizes suínas. Segundo a ANVISA, Agência de Vigilância Sanitária, o conceito de funcionalidade de

um ingrediente pode ser definido como um alimento rico em nutrientes capazes de atender as demandas metabólicas além do que é exigido, e que proporciona o aumento no desempenho dos animais. Além disso, o alimento funcional pode apresentar características eubióticas, ativando o metabolismo de bactérias benéficas no lúmen intestinal (PRECUP et al., 2019). Essa relação entre a fibra e a microbiota intestinal está intimamente relacionada com as propriedades físico-químicas intrínsecas das fibras e de acordo com a sua solubilidade (CAPUANO, 2017; DEEHAN et al., 2017).

Inúmeros oligossacarídeos conferem característica eubiótica, entre eles encontram-se os galactooligosacarídeos (GOS), frutoligosacarídeos (FOS) e inulina, todos agindo de forma específica no colón (VANDENPLAS et al., 2011). As bactérias benéficas, principalmente *Bifidobacterium* e *Lactobacillus* são estimuladas pelos FOS e inulina, favorecendo a redução de bactérias patogênicas. Além dessas bactérias, existem gêneros que são capazes de sintetizar AGVs que servem de substrato energético, para o hospedeiro e para outras bactérias presentes no intestino grosso, como por exemplo o gênero *Clostridium*. Essa produção de AGVs pode ser mediada através do consumo de fibras específicas, com características eubióticas (TOKACH et al., 2019).

As bactérias que foram beneficiadas com a inclusão da fibra, ao produzir os AGVs, estes caem na corrente sanguínea onde terão ação local, no fígado e em tecidos periféricos (KOH et al., 2016). Esses metabólitos têm relação com a sinalização de diferentes atividades fisiológicas, como as respostas imunológicas, ao estimular a expressão de fatores pró e anti-inflamatórios (GUO et al., 2020).

O estímulo à proliferação de bactérias benéficas através do consumo de fibras funcionais tem grande importância não só para a saúde da fêmea, mas também reflete no desenvolvimento da microbiota intestinal dos leitões (ZHOU et al., 2017). A transferência desses microrganismos pode ocorrer através do contato dos leitões com as fezes maternas ou via leite (ACHARD et al., 2019). Como exemplo, a família Christensenellaceae e concentrações elevadas de AGVs durante a lactação podem ser considerados padrões metabólicos importantes para promover a maturação do sistema imunológico dos leitões (LIU et al., 2019). Além disso, A família Christensenellaceae foi encontrada positivamente correlacionada com o elevado gasto energético, de acordo com o *status* metabólico de fêmeas suínas durante a lactação (LIU et al., 2015).

Tem sido cada vez mais relatado evidências de que a microbiota intestinal pode estar relacionada com o metabolismo das fêmeas suínas (XU et al., 2020; LI et al., 2020; HUANG et al., 2019; CHENG et al., 2018). A microbiota intestinal modulada pela inclusão de fibras funcionais na dieta desempenha um papel importante para a regulação metabólica durante o parto, principalmente quando se trata de resistência à insulina, comumente relatada em porcas nessa fase.

A resistência à insulina no parto deve-se pelo aumento dos níveis séricos de ácidos graxos livres, em resposta ao catabolismo intenso das fêmeas nessa fase (JI et al., 2017). Este distúrbio metabólico provoca uma resposta inflamatória basal, capaz de estimular a liberação de citocinas pró-inflamatórias e fator de necrose tecidual (TNF- α). Neste caso, a inclusão de fibra na dieta ao estimular a síntese de propionato, melhora a sensibilidade à insulina reduzindo a inflamação sistêmica (XU et al., 2018).

Entre os distúrbios metabólicos, também pode se destacar a obesidade das fêmeas suínas no parto. O aumento da relação entre os filos Firmicutes e Bacteroidetes aumentam notavelmente com o ganho de peso materno e a espessura de toucinho. Em mini-pigs obesos e em humanos com doença inflamatória intestinal, foi observada maior abundância do filo Actinobacteria (FRANK et al., 2007; PEDERSEN et al., 2013).

Tendo em vista os problemas causados pela obesidade, as fibras têm como vantagem a regulação da saciedade com o intuito de controlar, não só o estresse, mas também o consumo das fêmeas no terço final de gestação. Os AGVs produzidos são utilizados na gliconeogênese e na ativação de receptores que regulam a liberação de peptídeo semelhante ao glucagon (GLP-1) e o neuropeptídeo Y, todos responsáveis pelo controle da saciedade (TIAN et al., 2020).

Além disso, o uso de fibras funcionais pode auxiliar no melhor aproveitamento dos nutrientes dietéticos. A inulina, por exemplo, favorece a proliferação de microrganismos como as bactérias do gênero *Streptococcus*. Estas bactérias são capazes de degradar açúcares, levando a produção de ácido lático como produto final (VAN DEN BOGERT et al., 2013). Já as bactérias do gênero *Phascolarctobacterium*, são capazes de utilizar em maior proporção o succinato, substrato para produção de propionato, através da fermentação das fibras (ENGELS et al., 2016). As produções desses metabólitos, através da modulação da microbiota pela inclusão de inulina, são essenciais para a integridade intestinal (JARRETT e ASHWORTH, 2018; PATIL et al., 2020).

Portanto, os efeitos das fibras funcionais estão inteiramente relacionados com as suas características físico-químicas, modulando a microbiota para a produção específica de metabólitos desejáveis. A produção de AGVs a partir da degradação microbiana da fibra no intestino grosso tem ações sistêmicas, ao ativar a liberação de componentes imunológicos. Por isso, se deve compreender com detalhes a correlação entre esses processos fisiológicos, bem como a sua relação com a composição intrínseca das fibras funcionais.

3 CONSIDERAÇÕES GERAIS

A inclusão de fibras na dieta de matrizes suínas durante a gestação e lactação tem sofrido inúmeras restrições em decorrência do uso de fibras de procedências duvidosas, uma vez que a qualidade da fibra afeta diretamente no desempenho reprodutivo dos animais. Conhecer os componentes principais das fibras e suas propriedades físico-químicas são cada vez mais necessários, assim como elucidar os mecanismos de ação das fibras na microbiota intestinal e suas correlações com os metabólitos produzidos.

As fibras funcionais utilizadas no presente estudo podem conferir benefícios à saúde das matrizes suínas e de suas respectivas leitegadas, através da modulação microbiana e o estímulo à síntese de metabólitos que conferem melhora na integridade intestinal, além da ação imunomoduladora. Logo, a inclusão de fibras funcionais se torna uma alternativa cada vez mais efetiva na nutrição das matrizes suínas.

4 HIPÓTESE

A inclusão de fibras funcionais na dieta das matrizes suínas modula a microbiota intestinal, melhora a qualidade imunológica do leite e melhora o desempenho lactacional das fêmeas e de suas respectivas leitegadas.

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SECOND PART

ARTICLE

Journal of Animal Physiology and Animal Nutrition

**SOURCES OF FIBER IN GESTATION AND LACTATION AND THEIR
RELATIONSHIP WITH THE INTESTINAL MICROBIOTA AND THE
PERFORMANCE OF SOWS**

ABSTRACT

The aim of this study was to evaluate the inclusion of functional fiber in the diet and its impact on the intestinal microbiota, on the performance of sows and their respective litters, during gestation and lactation. Initially, 120 sows were selected, from 3 to 7 farrowing, distributed in random blocks. Sows received three diets during gestation and lactation: control treatment with 15% inclusion of soybean husk (CON), S25F treatment, control diet + 2.5% inclusion during gestation and 1.0% in lactation. partially soluble fiber and treatment (IF), control diet + inclusion of 2.5% during pregnancy and 1.0% insoluble fiber during lactation. 10 sows from each treatment were selected to evaluate blood parameters. In the 10 sows, volatile fatty acids (VFA), intestinal microbiota and pH were also evaluated in feces, at 111 days of gestation and at 16 days of lactation. At 16 days of lactation, immunoglobulins IgA and IgG were evaluated in the milk of sows. There was no significant difference between treatments regarding the performance of sows and piglets ($P > 0.05$). At 111 days of gestation, plasma urea levels were higher in CON compared to other treatments ($P < 0.10$). The percentage of IgG in CON milk was higher compared to S25F and IF and IgA was higher in S25F ($P < 0.05$). The levels of acetic and isobutyric acid were higher in CON compared to the other treatments ($P < 0.05$). In S25F, the highest abundance of the Euryarchaeota phylum was observed at 111 days of gestation ($P < 0.10$) and at 16 days of lactation ($P < 0.05$). At 111 days, the genus *Clostridium_IV* and *Methanobrevibacter* were more abundant in S25F ($P < 0.10$). In S25Fit was also observed lower relative abundance of the genus *Terrisporobacter* ($P < 0.05$). At 16 days of lactation, the relative abundances of the genus *Clostridium_IV* and *Clostridium_XIVa* were higher in CON ($P < 0.10$). The genus *Methanobrevibacter* was observed in greater abundance in S25F at 16 days of lactation ($P < 0.10$). It is concluded that the inclusion of functional fibers can improve the use of nutrients present in the diet, the immunological quality of milk, modulate the production of VFAs and alter the intestinal microbiota of the sows.

Keywords: Functional Nutrition, Intestinal Health, Litter, Microbiome

1. INTRODUCTION

The gestation phase is characterized as being the decisive phase for the swine production system, where important events occur for the performance of the subsequent phases. This period can be divided into three physiological phases, where in the final third of gestation there is greater metabolic activity (GAILLARD et al., 2019).

The peripartum can be considered the most critical phase for the health of the sows, since in this phase events such as the development of the mammary gland, the intense growth of fetuses and farrowing take place. When sows arrive at the peripartum with metabolic disorders due to poor body condition or nutritional failures, there is a metabolic overload at this stage, which can compromise lactational performance (LI et al., 2019). Insulin resistance and obesity are more notable disorders in the peripartum, which depress animal productive index by impairing the lactational performance of sows.

To reduce the impacts of metabolic disorders in the peripartum, nutritional strategies have been used throughout gestation and lactation. Among nutritional strategies, functional fibers have been gaining ground in the market, with positive results that improve the performance of sows. With eubiotic characteristics, the functional fibers, mainly lignocelluloses, have shown satisfactory results. Fiber, by stimulating the growth of beneficial bacteria in the gastrointestinal tract, aids in the synthesis of VFAs that will be metabolized by other microorganisms or will be absorbed by the host for energy synthesis (SHANG et al., 2021; HASHEMI et al., 2017; HUANG et al., 2019).

AGVs synthesized by microbial fermentation can aid in metabolic regulation, reducing systemic inflammation caused by insulin resistance. The main VFA involved in the regulation of insulin resistance is propionic acid, a product of microbial fermentation. Among the VFA produced, acetic acid is a substrate for the growth of bacteria that are beneficial to the host and, when metabolized, it can give rise to essential products for intestinal eubiosis. An intestinal microbiota in eubiosis is associated with better utilization of nutrients, as well as the production of metabolites that act on intestinal health (NIU et al., 2019).

Functional fibers have an immunomodulatory action, stimulating the synthesis of VFAs, which contribute to the control of local and systemic inflammatory responses. VFAs also regulate the production of immunoglobulins found in colostrum and milk from the sows, essential for the immunological development of piglets.

The use of functional fibers throughout the gestation has been used to ensure the health of sows, especially in peripartum and lactation. However, it is necessary to know the physicochemical properties and their effects on health, especially their effects on the intestinal microbiota of sows.

Therefore, the aim of this study was to evaluate the use of different eubiotic fibers throughout gestation and lactation their impacts on the peripartum, on the intestinal microbiota, on the health of females, VFAs, immunoglobulins, as well as on the lactational performance of sows and their respective litters.

2. MATERIAL AND METHODS

The project was submitted and approved by the Institutional Committee for Welfare and Ethics / Animal Protection of the Federal University of Minas Gerais (UFMG - CEUA), Brazil, identified by CEUA protocol 189/2019.

2.1 Location and Experimental Period

The experiment was conducted in the facilities of a multiplier farm with 700 sows, located in the Midwest region of Brazil, in the state of Goiás. The study was carried out from August 2020 to January 2021.

2.2 Animals and Buildings

A total of 120 multiparous sows, from three to seven parities of the same Topigs® Norsvin Line Z genetic, were monitored throughout gestation until the end of lactation. 90% of the sows were inseminated with semen from the male of breed Topigs® TLLLL Landrace and 10% with semen from the male line Topigs® TZZZZ Large White. The gestation sows selected did not present enteric, lameness or muscle problems, following the recommendations of Liu et al. (2019).

The gestation shed contained individual cages and had a trough-type feeder and a nebulization system for climate control. At 84 days of gestation, the sows were housed in a shed in collective pens until 110 days of gestation. Females were distributed in collective pens according to visual score and parities respecting the density of three to four sows per pen.

At 110 days of gestation, the sows were transferred to the maternity. The maternity rooms were equipped with calving cages (68 x 193 cm), containing a creep and a lamp to keep the ideal temperature for the piglets. The maternity was able to house 12 sows. All pens were made up of shell-type feeders and drinking fountain type of nipple, for both piglets and sows. The maternity sheds were composed of a Ductofan model air conditioning system, with refrigeration.

The sows remained in the maternity shed from 110 days of gestation until the weaning of the piglets, which was carried out, on average, on the 24th day of lactation. Temperature was measured throughout the experimental period, in at least two sheds in each production phase, using thermometers. The farm vaccination protocol was kept unchanged.

2.3 Experimental Design

The experiment was designed in randomized blocks, consisting of three treatments (Control-CON, Partially soluble fiber – S25F and Insoluble fiber - IF) with 40 multiparous females (three to seven parities) per treatment. Females were selected according to the order of parity (above three farrowing), body weight (BW) and backfat thickness (ET2) after insemination. Each female was considered an experimental unit, totaling 120 females.

2.4 Treatments e Experimental Diets

During the gestation period, the sows received two different experimental diets, but with the same level of fiber inclusion. From the day of insemination until 83 days, the sows received the following levels of fiber in the diet: the control treatment (CON), with inclusion of 15.0% of soybean hull; Treatment with 12.5% soybean hull plus 2.5% inclusion of partially soluble fiber (S25F); and the treatment, with 12.5% soybean hull plus 2.5% insoluble fiber (IF).

From 84 days until farrowing, sows received the following experimental diets: CON with 10.0% soybean hull inclusion; the S25F treatment with the inclusion of 8.5% soybean hull plus 2.5% partially soluble fiber; and the IF treatment with 8.5% soybean hull plus 2.5% insoluble fiber inclusion.

After farrowing, sows were fed diets with lower fiber inclusion during the lactation period, with CON treatment with 5.0% soybean hull, S25F with 4.0% soybean hull plus 1.0% soybean hull inclusion. partially soluble fiber and IF treatment with 4.0% soybean hull plus 1.0% inclusion of insoluble fiber.

The fibers evaluated in this study come from wood planted forests with the purpose of transforming wood into dietary fiber. IF is 100% insoluble, composed of 10% lignin. S25F is composed of 75% insoluble fiber and 25% soluble fiber arabinogalactan, with 30% lignin. The S25F and IF fibers are free from toxic components that are harmful to animal health. The composition of all experimental diets is described in the tables below.

Table 1. Chemical composition and energy values of fibers used in experimental diets

	Sources of fibers		
	Soybean Hull	25% Soluble Fiber ²	Insoluble Fiber
Dry matter, %	89.8	96.0	96.0
Crude fiber, %	32.9	65.0	70.0
NDF, %	58.1	5.0	5.0
ADF, %	46.1	70.0	75.0
ME kcal/kg	2,347	1,400	-
Lignin, %	2.1	30.0	10.0

¹Adapted: Sauvant and Tran, 2004; Rostagno et al., 2017. ²Insoluble fiber partially fermentable. NDF: Fiber in neutral detergent. ADF: Fiber in acid detergent. ME: Metabolizable Energy

Table 2. Composition of experimental diets provided to sows from artificial insemination to 83rd day of gestation

Ingredients	Composition Experimental Diets		
	CON	S25F	IF
Corn, %	63.40	63.40	63.40
Soybean meal, 46, %	17.20	17.20	17.20
Soybean hull, %	15.00	12.50	12.50
25% Soluble Fiber, %	-	2.50	-
Insoluble Fiber, %	-	-	2.50
Mastersorb premium, %	0.40	0.40	0.40
CNC-Gestation of sows, %	4.00	4.00	4.00
Total	100	100	100
Nutritional levels			
ME, kcal/kg	3,048.414	3,028.239	2,993.239
NE, kcal/kg	2,185.041	2,195.041	2,160.041
Crude protein, %	15.132	14.816	14.816
Non. dig. protein, %	2.41	2.31	2.31
Digestible lysine, %	0.737	0.724	0.724
Digestible threonine, %	0.528	0.520	0.520
Digestible tryptophan	0.152	0.150	0.150
Methionine, %	0.242	0.238	0.238
Leucine, %	1.148	0.148	0.148
Total leucine, %	0.565	0.555	0.555
Digestible Met+Cis, %	0.475	0.466	0.466
Digestible valine, %	0.634	0.624	0.624
Raw fat, %	3.123	3.068	3.068
Crude fiber, %	7.109	7.890	7.890
Soluble Fiber, %	1.35	1.30	1.305
Insoluble Fiber, %	17.29	17.72	17.73
Inert Fiber, %	4.97	5.97	6.47
Main fermented, %	5.33	5.29	5.20
Starch, %	43.74	43.6	43.6
Digestible phosphorus, %	0.382	0.379	0.379
Calcium (Ca), %	0.857	0.842	0.842
Sodium, %	0.217	0.216	0.216

Table 3. Composition of experimental diets provided to sows from the 84th day of gestation to farrowing

Ingredients	Composition Experimental Diets		
	CON	S25F	IF
Corn	62.10	61.10	61.10
Soybean meal, 46	17.00	17.00	17.00
Soybean hull	10.00	8.50	8.50
25% Soluble Fiber	-	2.50	-
Insoluble Fiber	-	-	2.50
Sugar	4.00	4.00	4.00
Degummed soybean oil	2.50	2.50	2.50
Mastersorb premium	0.40	0.40	0.40
CN-Prepartum 40	4.00	4.00	4.00
Total	100	100	100
Nutritional levels			
ME, kcal/kg	3,259.303	3,227.798	3,192.798
NE, kcal/kg	2,396.486	2,390.006	2,355.006
Crude protein, %	14.397	14.134	14.134
Non. Dig. protein, %	2.41	2.31	2.31
Digestible lysine, %	0.756	0.746	0.746
Digestible Leucine, %	0.482	0.475	0.475
Methionine, %	0.315	0.311	0.311
Leucine, %	1.126	1.117	1.117
Total isoleucine, %	0.537	0.528	0.528
Digestible Met-Cis, %	0.534	0.525	0.525
Digestible valine, %	0.605	0.595	0.595
Raw fat, %	5.369	5.309	5.309
Crude Fiber, %	5.431	6.532	6.532
Soluble fiber, %	1.35	1.30	1.305
Insoluble fiber, %	17.29	17.72	17.73
Inert fiber, %	4.97	5.97	6.47
Main fermented, %	5.33	5.29	5.20
Starch, %	43.74	43.6	43.6
Digestible phosphorus, %	0.388	0.386	0.386
Calcium (Ca), %	0.929	0.920	0.920
Sodium, %	0.155	0.154	0.154

Table 4. Composition of experimental diets provided to sows during the 24 days of lactation

Ingredients	Composition Experimental Diets		
	CON	S25F	IF
Corn	57.80	57.80	57.80
Soybean meal, 46	30.00	30.00	30.00
Soybean hull	5.00	4.00	4.00
25% Soluble Fiber	-	1.00	-
Insoluble Fiber	-	-	1.00
Degummed soybean oil	2.80	2.80	2.80
Mastersorb premium	0.40	0.40	0.40
CN-Lactation sows	4.00	4.00	4.00

Total	100	100	100
Nutritional levels			
ME, kcal/kg	3,377.867	3,369.797	3,355.797
NE, kcal/kg	2,414.444	2,418.444	2,404.444
Crude protein, %	19.517	19.391	19.391
Non dig. Protein, %	2.75	2.71	2.71
Digestible lysine, %	1.097	1.092	1.092
Digestible Threonine, %	0.835	0.832	0.832
Digestible tryptophan, %	0.232	0.231	0.231
Methionine, %	0.421	0.419	0.419
Leucine, %	1.526	1.526	1.526
Total isoleucine, %	0.768	0.764	0.764
Digestible Met-Cis, %	0.712	0.708	0.708
Digestible valine, %	0.859	0.855	0.855
Raw fat, %	5.610	5.588	5.588
Crude fiber, %	4.341	4.654	4.654
Soluble fiber, %	1.12	1.09	1.09
Insoluble fiber, %	12.68	12.85	12.86
Inert fiber, %	2.73	3.24	3.38
Main fermented, %	4.71	4.69	4.66
Starch, %	39.21	39.16	39.16
Digestible phosphorus, %	0.418	0.417	0.417
Calcium (Ca), %	0.929	0.923	0.923
Sodium, %	0.212	0.211	0.211

2.5 Animal Feed Management

Up to 83 days of gestation, the sows received gestational feed, following the feeding management described in the table below. From 84 days of gestation, the sows received a transitional diet (prepartum) until the time of parturition (Table 5).

Table 5. Feed management during the gestational period of sows supplemented with different sources of fiber

Period	In the morning	In the afternoon
Up to 35 days of gestation	2.2kg	-
36 to 83 days of gestation	2.0kg	-
84 days of gestation until Farrowing	1.3kg	1.3kg

At the maternity, females received lactation feed, following the recommendations of the Brazilian Tables of Poultry and Swine (ROSTAGNO et al., 2017). The lactational diet was gradually fed, increasing half a kilogram per treatment for the sows from the day of farrowing to the sixth postpartum day, up to 7.5 kg/day of lactation ration. The daily

amount of feed was divided into two main rations, according to the management carried by the farm.

The remaining ration was removed from the feeders and weighed in the morning of each day to evaluate the consumption of the sows during the lactation phase. Piglets received pelleted creep feeding from the fifth day of lactation until weaning. The lactating females and the piglets had free access to water.

2.6 Experimental Procedures

In the gestation barn, after insemination, all females were weighed on a scale with a precision of 1.0 kg and the backfat thickness was measured at point P2 (ET). The backfat thickness was measured at point P2 (6.5 cm from the dorsi-lumbar line and 6.5 cm from the last rib in the cranial direction), using the ALOKA® model SSD-500 ultrasonography equipment and 3-way linear transducer (5MHz UST 5011 model), following the methodology of Kim et al. (2015).

During the gestation period, 33 sows left the experiment due to problems with returning to heat and abortion. The 86 sows were weighed again at 110 days of gestation and then housed in the maternity barn, where the backfat thickness was measured at point P2 (ET2).

Blood samples were collected from the vein cava of 10 females of each treatment, two hours after morning feeding, at 111 days of gestation. In the afternoon, fresh feces were also collected from the same females used for the blood samples.

The sows' farrowings were watched and all piglets, including the stillborns, were counted and weighed before the feed to evaluate birth weight and the coefficient of variation of birth weight. Piglets from all treatments received the same routine newborn piglet care practices adopted on the farm. Up to 24 hours after birth, the piglets were weighed and equalized according to weight, with an average of 14 piglets remaining per litter, according to the number of viable teats. Equalization was performed between litters of the same treatment. The sows were weighed and ET2 was measured again 24 hours after parturition.

On the 16th day of lactation, blood samples were collected from the sows, respecting the fasting period of at least two hours. On the same day, milk and fresh feces samples were collected from the 10 sows of each treatment.

Blood samples were taken with the aid of a snare, using a hypodermic needle (40 x 1.6). The blood was deposited in two types of collection tubes, of which there were two 4 ml vacuum tubes containing EDTA K3 for plasma analysis and two 5 ml vacuum tubes with coagulation activator for serum analysis (TAN et al., 2015a).

After collection, the tubes containing EDTA K3 were poured for homogenization and placed in a thermos box with ice. Within a maximum interval of 40 minutes, the tubes were centrifuged under 1800 G rotation for 15 minutes to obtain blood plasma. After centrifugation, 1.5 ml of the supernatant was transferred to 2.0 ml microtubes and then stored in a freezer at -20°C.

Tubes without anticoagulant were immediately poured for homogenization and kept at room temperature for at least four hours and later centrifuged following the same procedures adopted for tubes with anticoagulant, to obtain serum. After centrifugation, 1.5 ml of the supernatant were transferred to 2.0 ml microtubes and then stored in a freezer at -20°C. Plasma and serum were sent to a specialized laboratory for biochemical analysis of blood compounds.

Fecal samples were collected fresh directly from the rectum of 10 sows, using nitrile gloves and plastic bags sanitized with 70% alcohol (CHENG et al., 2018). From the total collected feces, fractions of approximately 30 grams were removed, stored in two sterile conical bottom tubes of 50 ml (Falcon®) and four grams in a third conical tube. The stool samples contained in the third tube were diluted in distilled water to measure fecal pH using a calibrated pocket pHmeter (AKSO-AK95). Conical-bottom tubes with fecal samples containing around 30 grams, destined for VFA and MS, were immediately stored in a cooler containing reusable ice for transport to the freezer, where they were frozen at -20°C.

Small fractions of the total feces of approximately one gram were also removed, where they were put in three 2ml internal screw cryotubes. The samples contained in the cryotubes were immediately stored in a cylinder containing liquid nitrogen. The cylinder with liquid nitrogen containing fecal samples for analysis of the intestinal microbiota was sent to the NGS company, located in Piracicaba – SP, for analysis.

Before milk collection, the piglets were closed in the creepers, so that they would not have contact with the mother during milk collection. The sows' teats were sanitized with water and a paper towel. Soon after, 1 ml of carbetocin was injected intravenously in one

of the branches (lateral, intermediate, or medial) of the auricular tail vein of the sow. Milk was manually collected using sterile conical 50 ml tubes. Milk secretion was obtained from the mammary glands, interspersed between the thoracic, abdominal, and inguinal glands. After collection, the samples were frozen in a freezer at -20°C.

The evaluation of litter performance in the maternity was carried out by weighing the piglets at birth, weighing the litter up to 24 hours after birth and at weaning. Piglet mortality was measured based on the number of piglets born and the number of piglets weaned during the lactation period. One day before weaning, the sows were again weighed and ET2 measured.

2.7 Blood Tests

In the blood, urea levels were analyzed (Fixed-Time Kinetic Method), albumin (Bromocresol Colorimetric-Green Method), total proteins (Colorimetric-Biuret Method), total cholesterol (Enzymatic-Trinder Method) and its fractions (Colorimetric Method - Labtest) and triglycerides (Enzyme-Trinder Method).

2.8 Analysis of Fecal Samples

Microbial DNA extraction, genetic sequencing, dry matter (DM) content, fecal pH and analysis of volatile fatty acids (VFA) concentration were performed in feces.

For the extraction of microbial DNA, the Nucleic Acid Purification Kit MagMAX CORE (Thermo Fisher Scientific®) was used together with the complementary module for mechanical lysis MagMAX CORE (Thermo Fisher Scientific®), following the methodology described by the brand. After purification, nucleic acids were stored in a freezer at -20°C.

After extraction, confirmation of DNA integrity was verified by agarose gel electrophoresis (8%) and quantification by Nanodrop to obtain the concentration and absorbance ratio 260/280nm.

Primers (F:5-CCTACGGGNGGCWGCAG-3/R:5-GACTACHVGGGTATCTAATCC-3) (KLINDWORTH et al., 2013) were used for the amplification of the 16S rRNA gene, targeting the hypervaried region V3-V4. The PCR reaction was performed as follows:

	V3V4	
Reaction	1X	60X
ADN (storage)	1,0 µl	-
<i>Primer F</i> 10 µM	0,5 µl	5,0 µM
<i>Primer R</i> 10 µM	0,5 µl	5,0 µM
2X PCRBIO Ultra Mix	12,0 µl	120,0 µl
H2O	11,0 µl	100,0 µl
Total	25,0 µl	

For 16S V3V4:

- 95°C for 3 minutes 25 cycles
of:

- 95°C for 30 seconds
- 55°C for 30 seconds
- 72°C for 30 seconds

- 72°C for 5 minutes
- 4°C hold.

Purification of this PCR product was performed with Beckman AMPure XP Beads. After purification, ligation of the Illumina adapters was performed in a PCR reaction using 2.5 µl of purified PCR product, 2.5 µl of each index, 12.5 µl of 2X PCRBio Ultra Mix (PCR Biosystems) and 5.0 µl of ultrapure water for a final volume of 25.0 µl. This reaction also purified the samples with the beads.

The binding product of the adapters was normalized to the same concentration using the nanodrop. After normalization, a pool was made with the same volume of each sample and quantification by qPCR, to determine the pool concentration in nM. The kit used was the KAPA Library Quantification kit for the Illumina equipment.

After amplification, the amplicons were sequenced by the state-of-the-art sequencing platform, MiSeq Sequencing System (Illumina, Inc. San Diego, CA, USA). The entire procedure above was performed by the NGS Solutions Genomic Eirele Me laboratory, specialized in molecular biotechnology, located in Piracicaba - SP.

Initially, the primers were removed from the forward and reverse sequences using the “Trim Ends” tool. Then, using the “Merge Paired reads” tool, the forward and reverse sequences were aligned to form a single consensus sequence. The sequences were then filtered to remove low quality sequences (due to poor sequencing) and the “Workflows” tool was used to exclude sequences with less than 200 bp.

After filtering the sequences, the chimeras were removed using the UCHIME algorithm, where the sequences were compared with the SILVA database (QUAST et al., 2013). Then, the “16S Biodiversity” tool was used for taxonomic analysis of the microorganisms present in the samples. This tool attributes taxonomy to the sequences, comparing them with the Ribosomal Database Project (RDP) Classifier database (WANG

et al., 2007). The DNA sequences obtained by sequencing were analyzed using Geneious 10.2.3 bioinformatics software (KEARSE et al., 2012).

2.9 Determination of Volatile Fatty Acids

For the determination of short-chain volatile fatty acids, 1g of the sampled fecal content was weighed in a microtube and 1ml of distilled water was added. Afterwards, the solution was homogenized in a tube shaker and centrifuged at 15,000 G for 60 minutes at 4°C. Subsequently, the supernatant extract (± 0.4 ml) of each sample was transferred to chromatographic flasks, in which 100 μ l of 3:1 solution of 25% metaphosphoric acid with 98-100% formic acid and 50 μ l of solution of 100 mM 2-ethyl-butyric acid.

From this extract, 1 μ l was automatically injected by the injector system in a gas chromatograph (CG HP 7890A; Injector HP 7683B, Agilent Technologies) equipped with a capillary column HP-FFAP (1909F-112; 25 m; 0.32 mm; 0.5 μ m; JeW Agilent Technologies). The carrier gas used was H₂, maintained at a flow of 31.35 ml/min. The temperature of the injector and detector was 260°C and the total time of the chromatographic analysis was 16 and a half minutes, divided into three heating ramps: 80°C (1min.), 120°C (20°C/min.; 3 min.) and 205°C (10°C/min; 2 min.).

The acid concentration (mM) was determined based on an external calibration curve (FERREIRA et al., 2016). The analyzes were carried out at the Laboratory of Nutrition of Reproduction of the Luiz de Queiroz School of Agriculture –ESALQ of the Federal University of São Paulo (USP).

2.10 Determination of Fecal pH

After fecal collection from the sows, the samples were sent to the farm's insemination laboratory, where the fecal pH was checked. The stool samples contained in the 50 ml conical bottom tube were diluted 1:10 in distilled water (4 g:40 ml), homogenized with the aid of a vortex-type stirrer (Multifunctional Kasvi®). A pocket pHmeter calibrated and sanitized with paper towel, model AKSO-AK95, was inserted into the homogenized sample and then data annotation was performed.

2.11 Determination of Dry Matter of Fecal Samples

The evaluation of the dry matter (DM) content of the fecal samples was based on the INCT-CA G003/1 method of the National Institute of Science and Technology of Animal Science (DETMANN et al., 2012). The analysis of the DM content of the fecal

samples was carried out at the Animal Research Laboratory (LPA) of the Animal Science Department of the Federal University of Lavras, in Lavras-MG.

2.12 Milk Analysis

For the quantification of IgA and IgG immunoglobulins, milk samples were submitted to a non-reducing SDS PAGE. Total quantification was performed using a specific enzyme-linked immunosorbent kit (ELISA). The relative quantification of immunoglobulins was performed by band densitometry. The results generated by the product of the total quantification of IgG and IgA by the relative % of these were determined on the gel. The analysis was performed by an outsourced laboratory, located in Curitiba-PR.

2.13 Calculations and Statistical Analysis

The milk production of the sows was estimated according to the adapted equation of Noblet and Etienne (1989): Milk production (kg/day) = $[(0.718 \times \text{daily litter weight gain (g)} \times 100 - 4.9) \times \text{mean number of piglets}] / 0.19 / 1000$.

Data were tested for normality by the Shapiro-Wilk test, before analysis, and any variable that did not follow the normal distribution was transformed using the SAS RANK procedure (SAS INSTITUTE, 2009). Then, the data were submitted to analysis of variance, using the SAS statistical package through the F test ($P < 0.05$ and $P < 0.10$).

The results from fecal samples for sequencing the intestinal microbiota were submitted to the R studio program for data preparation and analysis statistics. Then, the data were submitted the Bioinformatics program Geneious® 10.2.3 software for the identification of microorganisms (KEARSE et al., 2012).

The relative abundance of microbial communities was verified by the Kruskal-Wallis test. For the alpha and beta diversity analyses, the R phyloseq package was used, based on the Shannon index for alpha-diversity analysis and the Bray-Curtis and PCoA Weighted Unifrac index for beta-diversity (McMURDIE and HOLMES, 2013).

3. RESULTS

During the experimental period, the sows were kept at barn temperature with an average of 23°C, maximum of 31.9°C, minimum of 21.4°C and relative humidity of 73.7%. The temperatures of each barn during the experimental period are described in table 6.

Table 6. Environmental parameters of the gestation barns, collective pens and maternity barns during the experimental period from August 2020 to January 2021

	Average T. (°C) ¹	Max. (°C) ²	Min. (°C) ³	Relative Humidity (%)
Gestation	23.1	32.8	21.1	63.1
Collective	22.7	32.0	20.9	-
Maternity	23.3	30.8	22.3	84.3

¹Average T.: average temperature. ²Max.: maximum temperature. ³Min.: minimum temperature

3.1 Performance of Sows

The performance results were not significant for the body condition of the sows after insemination, at 110 days of gestation and at weaning ($P>0.05$). The average feed intake did not express a significant difference among treatments during the gestation period and in the lactation phase ($P>0.05$). There was no influence of treatments on milk production of sows during lactation ($P>0.05$). The amounts of milk for each piglet were also statistically the same among treatments during lactation ($P>0.05$) (Table 7).

Tabela 7. Effect of experimental diets on performance of sows throughout the experimental period

	Treatments				<i>P-value</i>
	CON	S25F	IF	CV	
n	32	28	26		
<i>Post-insemination</i>					
Weight, kg¹	220.56	219.16	217.78	11.33	0.921
Backfat, mm	17.79	17.96	18.16	21.03	0.940
<i>111 days of gestation</i>					
Weight, kg	261.89	258.68	258.53	10.06	0.863
Backfat, mm	21.30	20.92	21.85	17.65	0.677
Feed intake, kg/day³	2.24	2.24	2.24	0.50	0.690
<i>Farrowing</i>					
Total born, n	16.81	18.61	17.27	20.82	0.112
Born alive, n	15.47	16.86	15.96	19.59	0.240
Stillborn, n	1.33	1.75	1.39	113.57	0.833

Mummified, n	0.13	0.18	0.43	271.90	0.189
CV birth, %	21.64	22.82	22.64	28.94	0.764
<i>24-hour post-farrowing</i>					
Weight, kg	243.45	239.84	242.25	9.91	0.848
Backfat, mm	18.62	17.58	18.18	19.64	0.514
<i>Weaning</i>					
Weight, kg	233.29	231.91	230.85	9.16	0.916
Backfat	16.93	16.73	16.98	17.97	0.950
<i>Lactation</i>					
Feed intake, kg/day⁴	6.152	6.237	6.106	8.13	0.524
Weight loss, kg	10.15	7.93	11.40	159.05	0.718
Weight loss, %	3.94	3.27	4.13	168.66	0.876
BF2 loss, mm⁵	1.69	0.57	1.2	252.24	0.367
BF2, %	8.44	4.16	4.47	254.29	0.528
MTindex⁶	2.45	2.53	2.64	19.58	0.382
Milk production kg/day	14.625	15.698	13.886	23.20	0.169
Milk/piglet, kg/day⁷	1.020	1.034	0.989	16.96	0.678

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber.¹ Body weight of sows. ³ Feed intake referring to the gestation phase of the sows that received a diet including different sources of fiber. ⁴ Feed intake referring to the lactation phase of the sows that received a diet including different sources of fiber. ⁵BF: backfat loss.⁶Mtindex: maternal transformation index of sow during the lactation period.⁷Average quantity of milk produced for each piglet. The numbers followed by the same letters did not differ from each other by the Tukey test at 5% probability.

3.2 Concentration of Immunoglobulins in Milk (IgG and IgA)

There was a significant difference in the percentages of IgG and IgA in the milk of the sows at 16 days of lactation ($P < 0.05$). The percentage of IgA was higher in the treatment with S25F inclusion, compared to the other treatments. The percentage of IgG was higher in the control treatment, and the other treatments showed lower percentages of the antibody. The IgG concentration was higher in the control treatment compared to the other treatments ($P < 0.027$) (Table 8).

Table 8. Effect of experimental diets on the concentration of immunoglobulins in the milk of sows, at 16 days of lactation

Variables	–	Treatments			CV	P-value
		CON	S25F	IF		
N		9	10	9		
IgA, %		0.76b	0.90a	0.89b	12.51	0.021
IgG, %		0.24a	0.10b	0.11b	72.59	0.021
IgA, µg/mL		586.16	723.23	854.02	40.38	0.150
IgG, µg/mL		169.44a	72.52b	103.28ab	72.08	0.027

Total	755.60	795.75	953.44	36.49	0.382
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CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

3.3 Litter Performance During the Lactation Period

Statistically, the treatments do not differ with respect to the performance of the piglets during the lactation period ($P > 0.05$), as demonstrate in Table 9.

Table 9. Effect of experimental diets on piglet performance during the lactation phase

	Treatments			CV	<i>P-value</i>
	CON	S25F	IF		
N	32	28	26		
Days of lactation	24.53	23.36	23.81	10.35	0.116
BW, kg¹	1.70	1.234	1.230	18.46	0.779
N piglets post-equalization	13.84	14.00	13.42	9.56	0.169
W24h, kg²	1.427	1.375	1.343	19.00	0.506
WL24h, kg³	19.72	19.18	18.10	20.63	0.332
N° weaning/litter	13.13	12.82	12.65	12.27	0.484
AWW, kg⁴	6.366	6.462	6.352	12.93	0.868
AWW litter, kg⁵	82.42	82.35	78.31	18.44	0.422
Mortality, n	0.70	1.18	0.83	149.49	0.447
DWG, kg⁶	0.215	0.224	0.216	16.20	0.562
DWGL, kg⁷	2.77	2.76	2.68	16.96	0.784

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. ¹BW: Birth weight. ²W24h: weight 24 hours post-farrowing. ³WL24h: litter weight 24 hours post-farrowing. ⁴AWW: average weaning weight. ⁵AWWlitter: average weaning weight of litters ⁶DWG: daily weight gain. ⁷DWGL: daily weight gain of litters. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

3.4 Blood Parameters

The S25F and IF diets tended to reduce plasma urea levels at 111 days of gestation compared to the CON treatment ($P < 0.085$). However, there was no significant difference in the other blood parameters evaluated at 111 days of gestation and 16 days of lactation ($P > 0.05$) (Table 10).

Table 10. Effect of treatments on the blood biochemical composition of sows

Variables	Treatments			CV	<i>P-value</i>
	CON	S25F	IF		
N	10	10	10		
111 days of gestation					
Urea, mg/dL	29.1 a	22.40 b	24.00 ab	28.14	0.085

Albumin, g/dL	3.09	2.98	2.93	15.30	0.758
Total Protein, g/dL	6.85	6.87	6.89	6.98	0.986
Total Cholesterol, mg/dL	59.60	54.30	52.40	16.58	0.280
LDL, mg/dL	23.16	20.44	23.82	47.27	0.704
HDL, mg/dL	17.50	14.90	14.70	41.59	0.598
VLDL, mg/dL	18.94	18.96	13.88	55.00	0.296
Triglycerides, mg/dL	94.70	94.80	69.40	55.00	0.296
<i>16 days of lactation</i>					
Urea, mg/dL	45.30	47.33	44.70	20.82	0.832
Albumin, g/dL	3.51	3.26	3.12	14.73	0.198
Total Protein, g/dL	5.51	5.71	5.64	6.03	0.448
Total Cholesterol, mg/dL	73.90	72.25	68.40	17.84	0.632
LDL, mg/dL	29.94	27.95	27.38	32.36	0.822
HDL, mg/dL	38.30	40.25	36.40	21.49	0.629
VLDL, mg/dL	5.66	4.05	4.62	48.59	0.171
Triglycerides, mg/dL	28.30	20.25	23.10	48.59	0.171

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. ¹LDL: low-density lipoprotein. ²HDL: high-density lipoprotein. ³VLDL: very low-density lipoprotein. The means followed by the same letter did not differ from each other at the 5% probability level by the Tukey test

3.5 Fecal Concentration of Volatiles Acid Fatties

The highest concentrated treatment of acetic acid in the feces of the sows at 111 days of gestation, compared with the concentration of the other concentrates ($P < 0.047$). The inclusion of S25F in the diet had a lower concentration of acetic acid compared to the control treatment and a similar result to the treatment with IF inclusion. The treatment also concentrated isobutyric acid compared to the other treatments ($P < 0.039$). The other treatments showed parallel results with each other. At 16 days of lactation, there was no significant difference among treatments regarding the concentration of VFAs in the feces of the sows ($P > 0.05$) (Table 11).

Table 11. Effect of treatments in concentration of fecal volatile fatty acids, in mM, of sows, at 111 days of gestation and 16 days of lactation

Variables	Treatments			CV	P-value
	CON	S25F	IF		
N	10	10	9		
<i>111 days of gestation</i>					
Acetic	57.98a	45.63b	53.81ab	22.78	0.047
Butyric	7.29	5.59	7.12	34.93	0.184

Isobutyric	1.72a	1.60ab	1.2ab	30.69	0.039
Propionic	9.23	7.81	6.71	68.45	0.520
Valeric	1.92	1.69	1.69	23.55	0.375
Isovaleric	3.43	3.17	3.07	20.49	0.483
Total	81.57	65.50	73.60	24.07	0.112

16 days of lactation

Acetic	74.65	75.83	71.01	22.39	0.813
Butyric	10.37	11.21	10.24	31.27	0.789
Isobutyric	2.32	2.17	2.24	24.75	0.918
Propionic	20.19	22.58	19.97	28.97	0.577
Valeric	2.33	2.23	2.15	24.58	0.803
Isovaleric	3.74	3.62	3.52	24.20	0.767
Total	113.60	117.64	109.13	23.08	0.783

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

3.6 Fecal pH and Dry Matter

There was no significant difference among treatments in relation to the fecal pH of the sows, at 111 days of gestation and at 16 days of lactation ($P>0.05$). The dry matter content of feces of sows was not different among treatments, at 111 days of gestation and at 16 days of lactation ($P>0.05$) (Table 12).

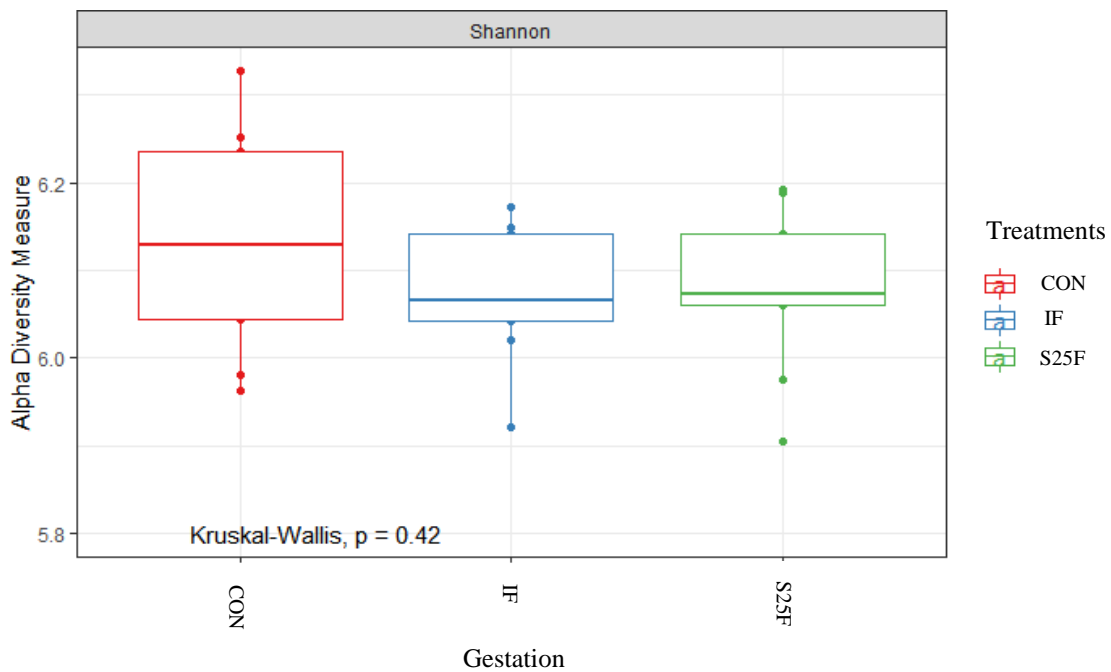
Tabela 12. Effect of treatments on fecal pH and dry matter (DM) of sows, at 111 days of gestation and 16 days of lactation

Variables	Treatments			CV	P-value
	CON	S25F	IF		
N samples	10	10	8		
<i>111 days of gestation</i>					
Fecal pH	7.79	7.75	7.74	4.69	0.948
DM	36.86	37.13	35.00	10.96	0.493
<i>16 days of lactation</i>					
Fecal pH	7.07	7.02	7.20	6.79	0.602
DM	30.43	29.99	29.69	10.84	0.902

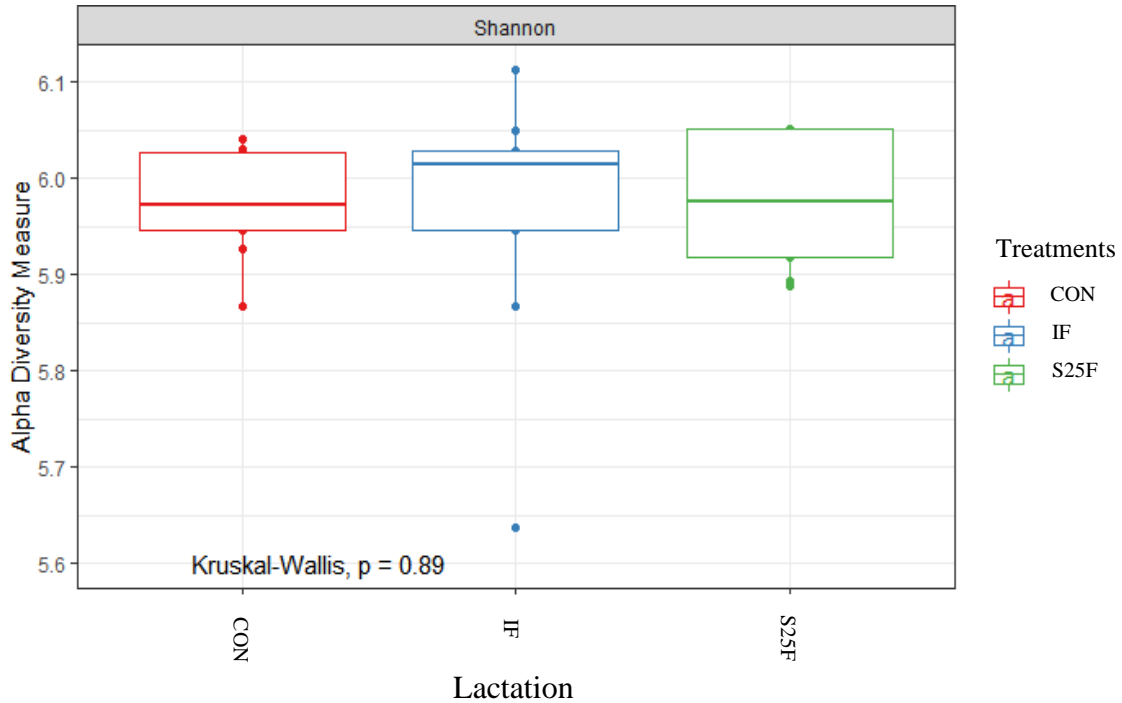
CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

3.7 Intestinal Microbiota of Sows

To evaluate the structure of the microbial community present in each treatment, the Shannon index was used. At 111 days of gestation, Shannon index values were 6.1, 6.08 and 6.07 for the control treatments, S25F and IF, respectively. At 16 days of lactation the values were 5.97, 5.97 and 5.96 for the Control, S25F and IF treatments, respectively. There was no difference among treatments at 111 days of gestation, nor at 16 days of lactation ($P>0.05$) (Graphic 1 and 2).

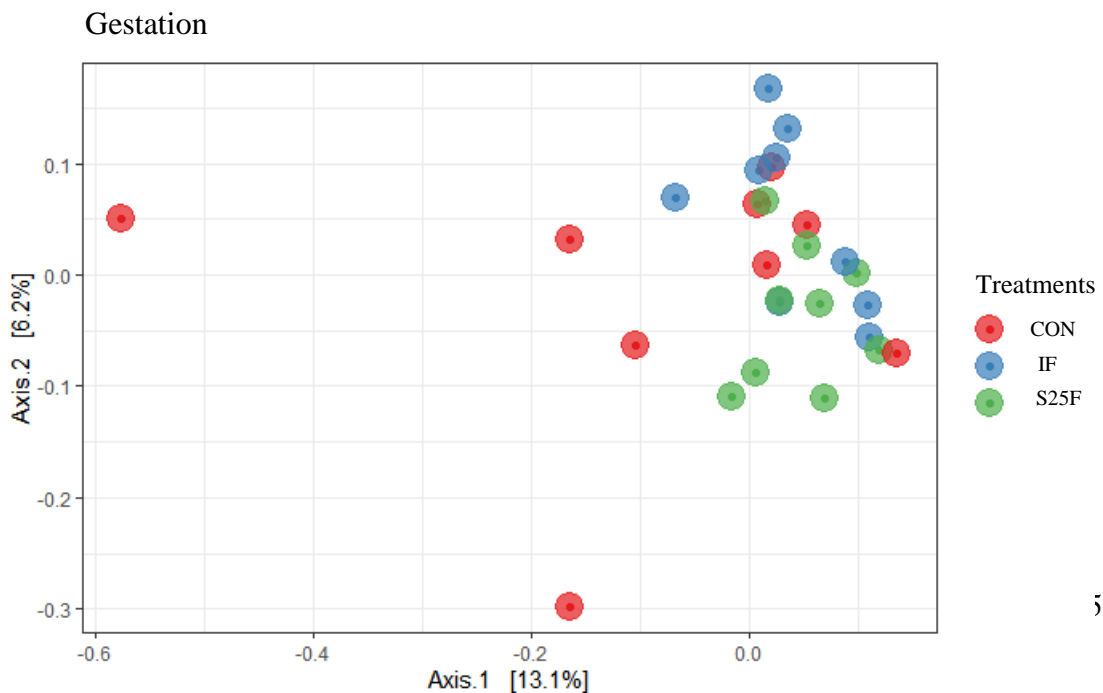


Graphic 1. Effect of alpha-diversity treatments on the fecal microbial community of sows provided by the Shannon index at 111 days of gestation

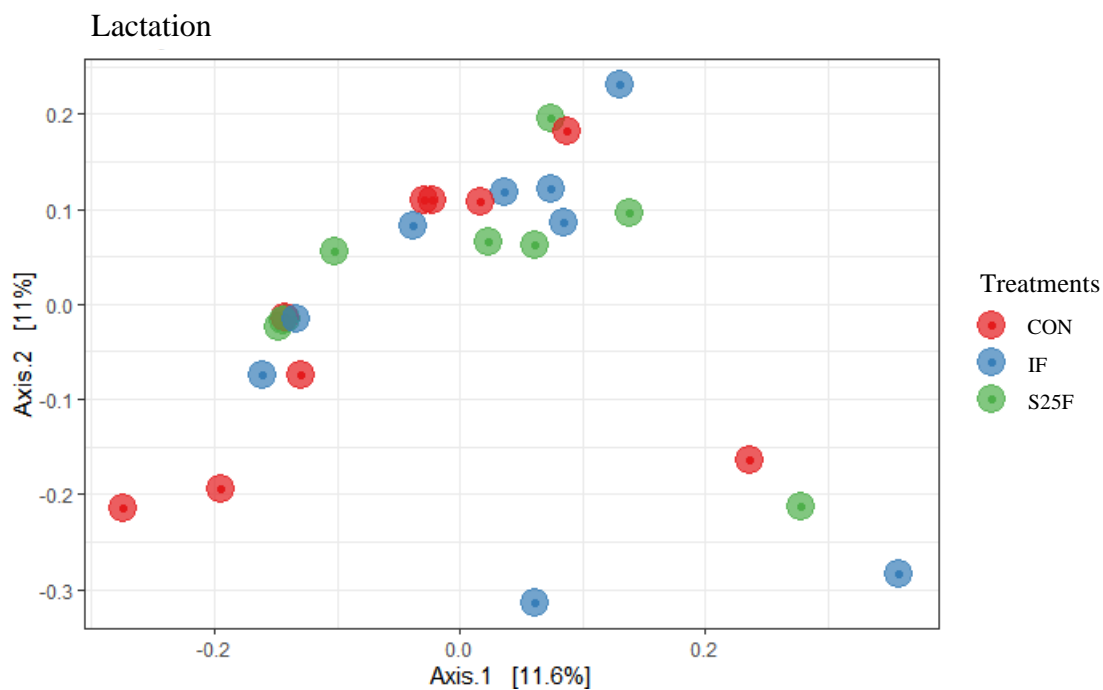


Graphic 2. Effect of alpha-diversity treatments on the fecal microbial community of sows provided by the Shannon index at 16 days of lactation

The PCoA results in weighted Unifrac distance revealed distinct displacement patterns, without grouping of individuals at 111 days of gestation and at 16 days of lactation (Graphs 3 and 4). Therefore, there was no difference among treatments ($P > 0.05$).

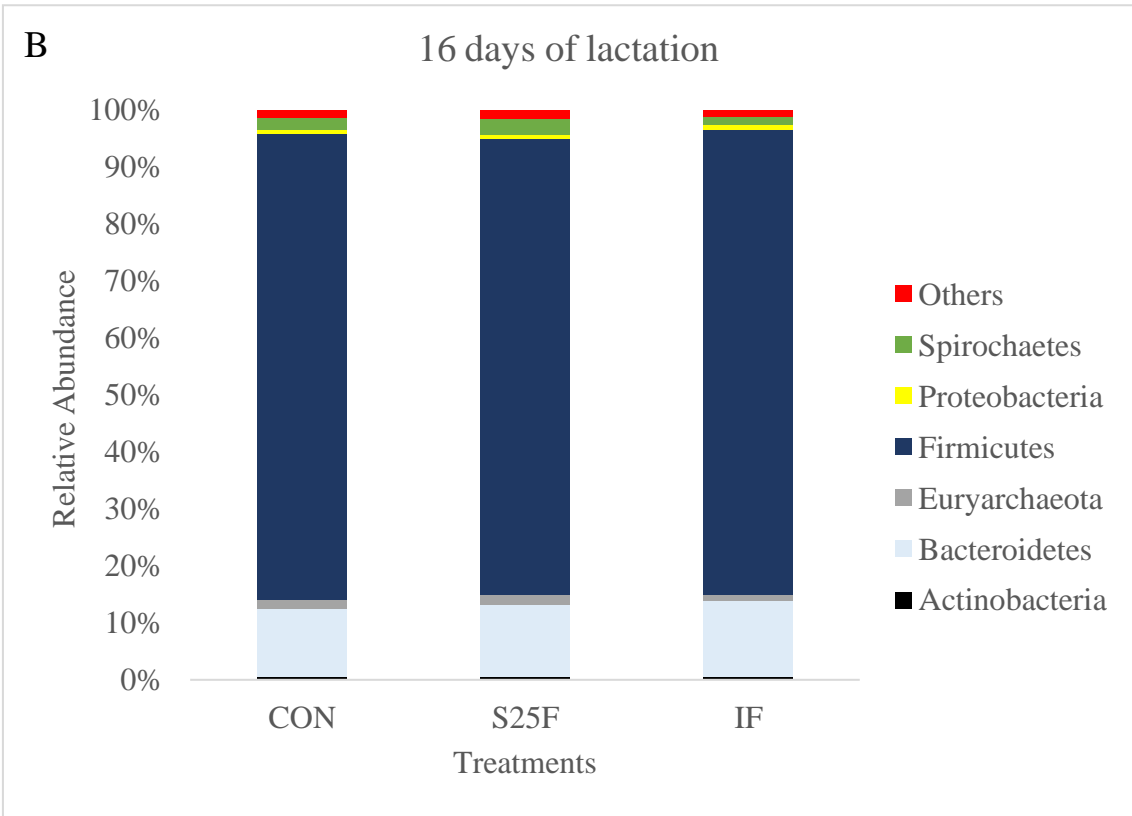
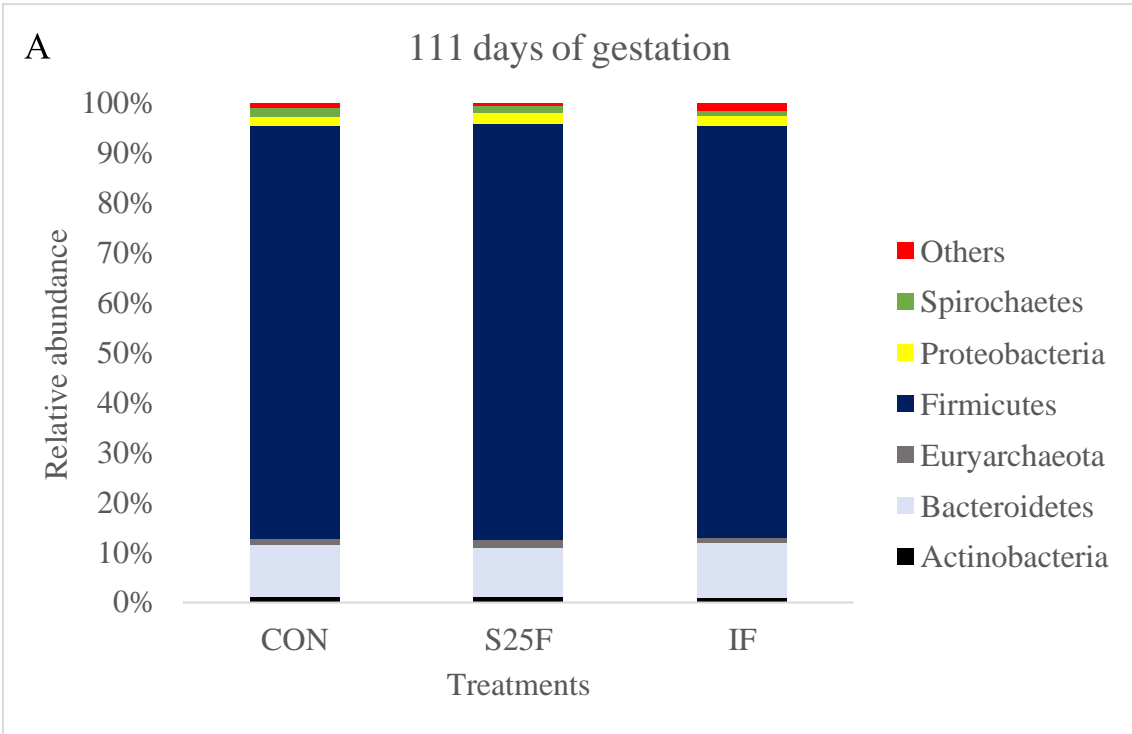


Graphic 3. PCoA profile based on the weighted Unifrac distance of the OTUs of each fecal sample from sows fed with inclusion of different fiber of sources at 111 days of gestation



Graphic 4. PCoA profile based on the weighted Unifrac distance of the OTUs of each fecal sample from sows fed with inclusion of different fiber of sources at 16 days of lactation

Six phyla and 53 different genera were identified in all treatments. The 25 most frequent and abundant genera are represented in Graph 6 and 7.



Graph 5. Effect of treatments on the relative abundance (%) of the phylum level observed in the intestinal microbiota of sows at 111 days of gestation (A) and at 16 days of lactation (B)

In the sequencing of the 16S gene, specifically in the hypervariable region V3-V4, the phyla Firmicutes, Bacteroidetes, Proteobacteria, Actinobacteria and Spirochaetes were identified in all treatments. All the phyla mentioned above were found in the feces of the sows at 111 days of gestation and at 16 days of lactation (Graph 5). The Firmicutes phylum (CON = 82.8%, S5F = 83.2% and IF = 82.4%) was identified in the highest percentage, followed by Bacteroidetes (CON = 10.4%, S25F = 9.9 and IF = 11.1) and Proteobacteria (CON = 1.9%, S25F = 2.2% and IF = 2.0%) in all treatments, at 111 days of gestation. At 16 days of lactation, the most predominant phyla were Firmicutes (CON = 81.9%, S25F = 80.4% and IF = 81.7%), Bacteroidetes (CON = 11.9%, S25F = 12, 4% and IF = 13.4%) and Spirochaetes (CON = 2.1%, S25F = 2.8% and IF = 1.4%), in all treatments.

It was observed a trend to a higher relative abundance of Euryarchaeota in the S25F treatment (1.6%), compared to the other treatments (CON = 1.2% and IF = 1.0%) at 111 days of gestation ($P < 0.088$). With respect to the other phyla, there was no significant difference among treatments at 111 days of gestation ($P > 0.05$) (Table 13).

Table 13. Effect of treatments on relative abundance (%) at phylum level observed in the intestinal microbiota of sows at 111 days of gestation

Phylum (111d gestation)	Treatments			CV	P-value
	CON	S25F	IF		
n	9	9	9		
Actinobacteria	1.078	1.167	0.878	59.84	0.823
Bacteroidetes	10.444	9.889	11.111	38.83	0.828
Euryarchaeota	1.244	1.622	0.989	45.56	0.088
Firmicutes	82.778	83.222	82.444	4.88	0.925
Proteobacteria	1.889	2.222	1.978	42.58	0.591
Spirochaetes	1.622	1.300	1.189	52.65	0.506
Others	0.944	0.578	1.411	89.71	0.164

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

At 16 days of lactation there was a significant difference among treatments Euryarchaeota abundance ($P < 0.047$). Treatment S25F (1.8%) More current relative than the other treatments (CON = 1.6% and IF = 1.0%). In the IF treatment, a lower prevalence of the phylum Euryarchaeota was observed (Table 14).

Table 14. Effect of treatments on relative abundance (%) at phylum level observed in the intestinal microbiota of sows at 16 days of lactation

Phylum (16d lactation)	Treatments			CV	P-value
	CON	S25F	IF		

n	9	9	9		
Actinobacteria	0.544	0.511	0.467	82.34	0.918
Bacteroidetes	11.889	12.444	13.444	32.61	0.733
Euryarchaeota	1.556 ab	1.778 a	1.011 b	46.50	0.047
Firmicutes	81.889	80.444	81.667	5.98	0.807
Proteobacteria	0.689	0.678	0.878	73.06	0.383
Spirochaetes	2.111	2.778	1.433	80.54	0.336
Others	1.322	1.367	1.100	61.73	0.954

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

At the genus level, bacteria such as *Clostridium sensu stricto*, *Romboutsia*, *Turicibacter*, *Terrisporobacter* and *Prevotella* were the most abundant in treatments at 111 days of gestation. At 16 days of lactation, the most abundant bacteria were *Clostridium sensu stricto*, *Lactobacillus*, *Romboutsia*, *Turicibacter*, *Terrisporobacter* and *Prevotella*.

Trends were observed regarding the relative abundance of the genus *Clostridium IV* (CON = 0.1%, S25F = 0.6% and IF = 0.3%) and *Methanobrevibacter* (CON = 1.2%, S25F = 1.6% and IF = 1.0%) at 111 days of gestation ($P < 0.10$). These two genera were higher in the S25F treatment compared to the other treatments. There was a significant difference in the relative abundance of the genus *Terrisporobacter* ($P < 0.017$), since the S25F treatment (6.9%) showed lower abundance of the genus at 111 days of gestation, compared to the other treatments (CON = 8.2 and IF = 8.1). The abundance of unspecified genera was higher in the IF treatment compared to the other treatments ($P < 0.001$). There was no significant effect of treatments on the relative abundance of genera ($P > 0.05$) at 111 days of gestation (Graph 6 and table 15).

Table 15. Effect of treatments on relative abundance (%) at genus level observed in the intestinal microbiota of sows at 111 days of gestation

genus (111d gestation)	Treatments			CV	P-value
	CON	S25F	IF		
n	9	9	9		
<i>Acidaminobacter</i>	1.53	1.93	0.84	92.72	0.166
<i>Anaerobacter</i>	1.33	1.33	1.67	35.06	0.273
<i>Barnesiella</i>	1.78	2.08	2.11	44.85	0.881
<i>Christensenella</i>	1.41	0.97	1.00	68.95	0.529
<i>Clostridium IV</i>	0.11	0.60	0.31	125.11	0.086
<i>Clostridium sensu stricto</i>	27.11	30.56	29.78	18.37	0.564
<i>Clostridium XIVa</i>	2.44	2.44	2.33	36.90	0.950
<i>Escherichia/Shigella</i>	0.74	0.76	0.88	104.80	0.865
<i>Falsiporphyromonas</i>	0.21	0.40	0.21	158.26	0.554

<i>Intestinimonas</i>	0.50	0.73	0.32	86.42	0.410
<i>Lachnospiracea incertae</i>	2.22	2.00	1.78	39.22	0.607
<i>Lactobacillus</i>	3.78	1.62	0.78	140.87	0.308
<i>Methanobrevibacter</i>	1.21	1.62	0.99	47.67	0.088
<i>Oscillibacter</i>	0.89	0.94	1.11	49.43	0.505
<i>Paraprevotella</i>	0.54	0.11	0.21	228.88	0.529
<i>Phascolarctobacterium</i>	0.33	0.51	0.82	98.88	0.356
<i>Prevotella</i>	3.22	3.11	4.11	50.62	0.435
<i>Romboutsia</i>	5.22	6.44	5.33	24.47	0.116
<i>Ruminococcus</i>	0.11	0.08	0.00	366.30	0.594
<i>Sporobacter</i>	1.43	1.64	1.67	37.04	0.617
<i>Streptococcus</i>	4.00	2.22	2.89	72.76	0.221
<i>Terrisporobacter</i>	8.22 a	6.89 b	8.11 a	15.03	0.017
<i>Treponema</i>	1.44	1.21	1.11	68.53	0.603
<i>Turicibacter</i>	9.11	10.00	10.22	26.20	0.640
<i>Others</i>	18.96 b	16.99 b	20.33 a	11.13	0.001

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test

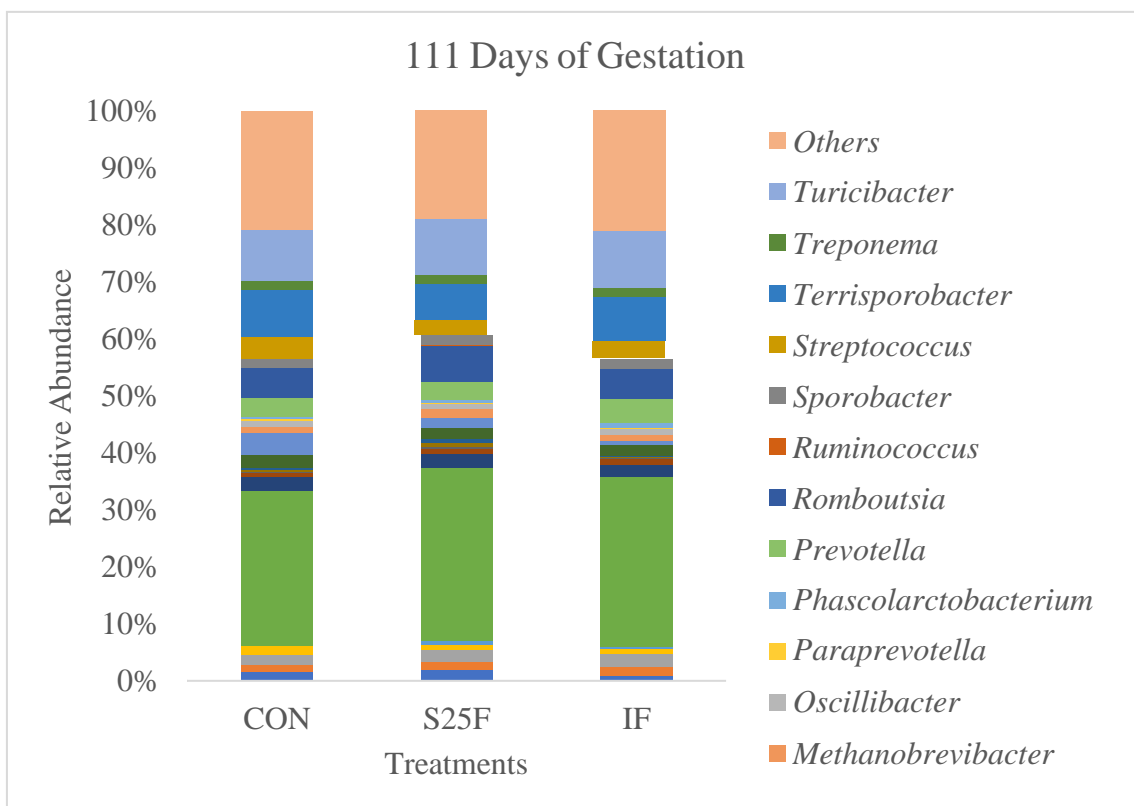
At 16 days of lactation, the results were biased ($P < 0.10$) for the relative abundance of the genus *Clostridium IV* (CON = 1.0%, S25F = 0.8% and IF = 0.6%), *Clostridium XIVa* (CON = 2.0%, S25F = 1.7% and IF = 1.3%) and *Metanobrevibacter* (CON = 1.6%, S25F = 1.8% and IF = 1.0%). The genus *Clostridium IV* and *Clostridium XIVa* expressed greater abundance in the control treatment, contrasting with the other treatments. The genus *Methanobrevibacter* was more abundant in the S25F treatment. The other genera did not express significant results ($P > 0.05$) (Graphic 7 and table 16).

Table 16. Effect of treatments on relative abundance (%) at genus level observed in the intestinal microbiota of sows at 16 days of lactation

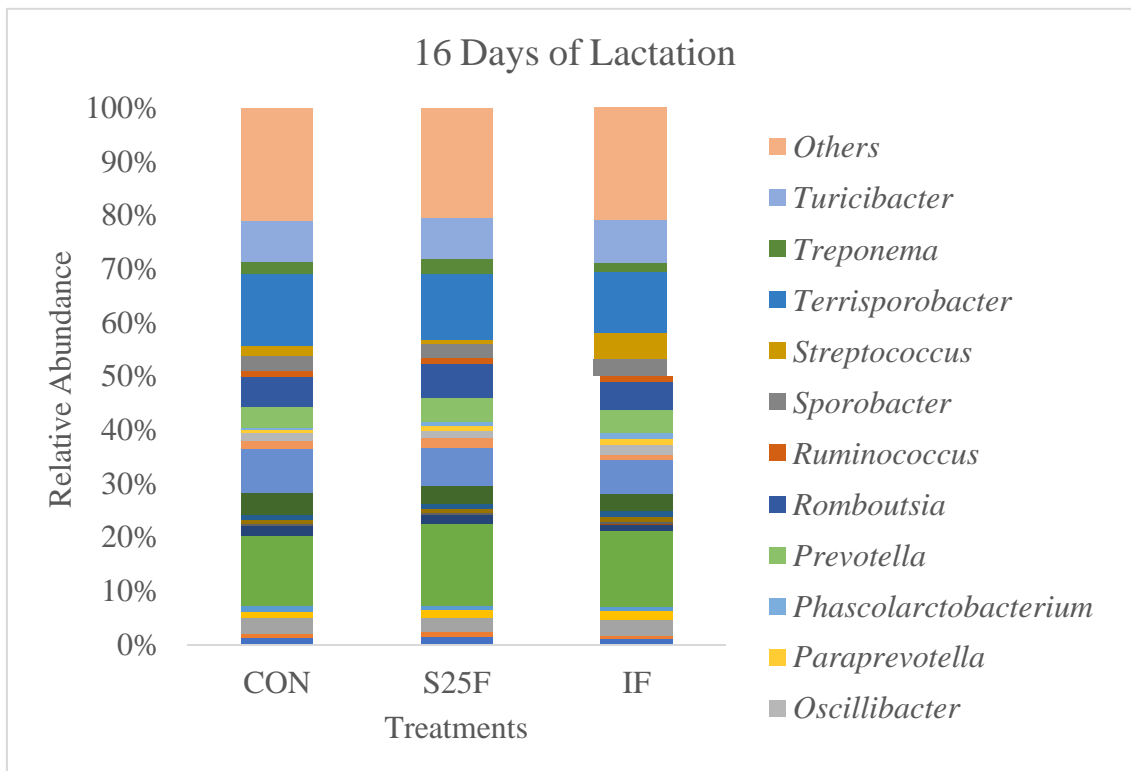
genus (16d lactation)	Treatments			CV	P-value
	CON	S25F	IF		
n	9	9	9		
<i>Acidaminobacter</i>	1.41	1.40	1.06	66.44	0.699
<i>Anaerobacter</i>	0.67	0.96	0.72	56.79	0.265
<i>Barnesiella</i>	2.89	2.78	3.00	37.59	0.941
<i>Christensenella</i>	1.31	1.40	1.64	56.75	0.681
<i>Clostridium IV</i>	0.96	0.83	0.60	38.86	0.083
<i>Clostridium sensu stricto</i>	13.00	15.00	14.11	25.72	0.518
<i>Clostridium XIVa</i>	2.00	1.67	1.33	44.03	0.099
<i>Escherichia/Shigella</i>	0.00	0.00	0.11	519.62	0.368
<i>Falsiporphyrromonas</i>	0.37	0.22	0.37	135.19	0.774
<i>Intestinimonas</i>	0.81	0.90	1.02	33.94	0.646

<i>Lachnospiracea incertae</i>	4.33	3.67	3.22	36.01	0.218
<i>Lactobacillus</i>	8.11	6.56	6.30	91.69	0.741
<i>Methanobrevibacter</i>	1.56	1.77	1.01	46.81	0.070
<i>Oscillibacter</i>	1.56	1.44	1.89	34.66	0.256
<i>Paraprevotella</i>	0.34	0.82	0.93	94.84	0.110
<i>Phascolarctobacterium</i>	0.56	0.78	1.14	98.97	0.664
<i>Prevotella</i>	3.78	4.22	4.22	41.92	0.921
<i>Romboutsia</i>	5.67	6.11	5.44	33.68	0.779
<i>Ruminococcus</i>	1.03	1.02	1.08	105.33	0.903
<i>Sporobacter</i>	2.78	2.56	2.44	37.46	0.845
<i>Streptococcus</i>	1.99	2.89	5.22	244.85	0.586
<i>Terrisporobacter</i>	13.44	12.00	11.56	26.70	0.462
<i>Treponema</i>	2.10	2.78	1.43	79.72	0.389
<i>Turicibacter</i>	7.67	7.56	8.11	41.48	0.933
<i>Others</i>	18.17	17.97	17.09	14.05	0.454

CON – control treatment; S25F – 25% soluble fiber; IF – insoluble fiber. The means followed by the same letters did not differ from each other at the 5% probability level by the Tukey test



Graphic 6. Effect of treatments on the relative abundance at the genus level of the intestinal microbiota of sows at 111 days of gestation



Graphic 7. Effect of treatments on the relative abundance at the genus level of the intestinal microbiota of sows at 16 days of lactation

4. DISCUSSION

The objective of this study was to evaluate the use of functional fibers in the diet of sows during gestation and lactation and its effects on the intestinal microbiota and performance during the gestation and lactation phase. For this, experimental diets were formulated with the inclusion of 2.5% of functional fiber in the gestational phase and 1% of the same fibers in the lactational phase of the sows.

During the experimental period, the sows remained 97% of the days exposed to temperatures above 26°C, as can be seen in Table 18. Considering all the barns, in 74% of the days as sows were exposed to temperatures above 30°C, with an average air humidity of 73.5%. Thus, it can be stated that during the experimental period the animals in this study were exposed to constant periods of heat stress (LUCY and SAFRANSKI, 2017).

As seen in Table 7, the inclusion of defined fiber in the diet of sows did not result in significant changes in body condition during the gestational phase. The parities chosen to evaluate the effects of fibers may have contributed to the result. Younger sows, from

first and second parturition, lose body weight more significantly than sows above three parturitions (KIM et al., 2015).

However, in studies carried out by Shi et al. (2021) found lower backfat thickness at 107 days of gestation in the treatment that received high levels of fiber compared to the control treatment. The same result was found by Zhou et al. (2017) by including 1.5% inulin in the diet of gestation sows. Szuba-trznadel et al. (2017) reported significant losses during lactation of sows that received a diet including 0.05% mannanooligosaccharides (MOS).

The inclusion of fiber in the diet did not change in the feed intake during the sows' lactational phase. Different results found by Shang et al. (2019), where feed intake was higher during lactation by including 10% beet pulp in the lactational diet, compared to the control treatment, however, it did not differ from the treatment with inclusion of wheat bran. Consumption during lactation is of great importance for success at this stage, as it ensures greater intake of nutrients for the female's basal metabolism and for milk production (THEIL et al., 2014). In this case, a beet pulp increased feed intake, improving the insulin sensitivity of sows (TAN et al., 2016c).

During a gestation phase the sows were under food restriction, which was followed by the recommendations of the genetics company of the sows. Thus, the sows consumed what was offered during gestation. However, in the lactation period, feed intake was below (± 6 kg / day) than what was offered in all treatments (7.5kg / day). This low intake may be related to heat stress, given the high temperatures they were exposed to in the experimental period. Temperature is directly related to the reproductive and productive performance of production animals. The reduction in feed intake is considered a natural mechanism to slow down the metabolism of animals exposed to high temperatures (MUNS et al., 2014; WILLIAMS et al., 2013).

Although feed intake was lower than that provided in all treatments, the percentage of weight loss during lactation was less than 10% for multiparous sows, which is the percentage of limit loss found by Cozannet et al. (2018). In this meta-analysis, the author found some works that revealed considerable decrease in the performance of litters of sows with more than 10% of body condition losses. This result highlights the importance of using good quality fiber in lactation diets, as according to studies carried out by Hu (2019), excessive weight loss during lactation can delay the weaning-estrus

interval and impair subsequent reproductive performance. According to Hoving et al. (2012), excessive weight loss during lactation can affect female productivity, with a decrease in the ovulation rate and a late return to heat. The loss and excess weight are harmful to the breeding females of the herd, causing the depreciation of animal productivity indices and economic deficit (THAKER and BILKEI, 2005; KNAUER, 2018).

Another factor that could explain the low efficiency of the fibers evaluated is the bromatological profile and the high inclusion of soybean hulls in the S25F and IF treatments in the present study. The physical and chemical properties of fiber may be associated with the use of nutrients present in the diet (HAMAKER et al., 2014). Intrinsic factors such as solubility, fermentation capacity and retention time can affect the use of fiber in non-ruminant animals (OLIVIERO et al., 2009; JHA et al., 2019).

It was not possible to find a significant difference in the production of sows that received different dietary sources of fiber. Milk production in all treatments at 16 days was on average 14.73 kg / day, a value higher than the findings by Martins et al. (2007), Renaudeau et al. (2003) and Quiniou and Noblet (1999), where the authors evaluated the colostrum and milk production of sows in heat stress conditions. Genetics has contributed to greater production, which is associated with a greater number of piglets born in the last 10 years (QUESNEL et al., 2015).

The immunoglobulin concentration is a parameter of great relevance when it comes to the quality of colostrum and milk because they show greater immunological protection, both maternal and offspring. Mainly in pigs, a species dependent on the transfer of antibodies by colostrum and milk (NECHVATALOVA et al., 2011). According to the review written by Tian et al. (2020), nutrition can change the composition of colostrum and milk from sows, especially with the dietary inclusion of functional fibers. However, studies on the effects of fibers on immunoglobulin secretion are inconsistent (LOISEL et al., 2013; SHANG et al., 2019).

In the present study, the percentage of IgG in milk at 16 days of lactation was higher in the control treatment compared to treatments with inclusion of S25F and IF. The IgG present in colostrum is absorbed by piglets up to 24 hours after parturition. From that moment, piglets lose the ability to absorb the immunoglobulins present in the milk (QUESNEL and FARMER, 2019).

IgG is an immunoglobulin with systemic action, and its high levels in the blood can be a sign of an immune response to infectious conditions (SCHAREK et al., 2005). Furthermore, heat stress can induce gastrointestinal disturbs, through the imbalance of inflammatory responses and lymphocyte resistance to glucocorticoids (GIMSA et al., 2018). Taking this fact into account, the inclusion of S25F fiber may have contributed to the reduction of some stress factors, different from the control treatment, where the immune response was more accentuated. The extra IgG produced by the immune system could be observed in higher concentrations in milk (SCHAREK et al., 2005; TIZARD, 2014).

In the S25F treatment there was 25% arabinogalactan, which may have differentiated from the other treatments. Some polysaccharides can stimulate immune cells, as well as cytokines present in organisms. Arabinogalactans are polysaccharides able to stimulate T cells and macrophages present in the intestinal mucosa to produce immunoglobulins (TINGIRIKARI et al., 2018). They also stimulate the release of pro and anti-inflammatory cytokines for the regulation of the immune response (CHOI et al., 2005).

In the present study, a higher percentage of IgA was observed in the treatment with S25F inclusion, when compared to the percentages of the other treatments. This result reveals an ability of the S25F fiber to stimulate the production of antibodies to protection of the female and transfer sIgA to the piglets. In a study conducted by Gourbeyre et al. (2012) also observed higher levels of IgA in the milk of rats supplemented with a mixture of galactooligosaccharide and inulin, eight weeks after parturition. Different from the results found by Le Bourgot et al. (2014), where the inclusion of short chain fructooligosaccharide in the diet of sows during gestation and lactation reduced the concentration of sIgA in milk at 6 days of lactation.

IgA present in milk, like IgG, is not absorbed by the gastrointestinal tract after 24 hours. However, the IgA when ingested by piglets is fixed in the intestinal mucosa and has an antimicrobial action against pathogenic microorganisms (KAFKOVA et al., 2020). sIgA can neutralize infectious agents, binding to the membrane receptors. With inoperative receptors, microorganisms cannot bind to enterocytes, passing directly along with the indigestible portion through the intestinal lumen (TIZARD, 2014). Furthermore, in pigs, IgA is found at a higher concentration in milk than in colostrum, compared to

other immunoglobulins (BUTLER et al., 2017; TIZARD, 2014). When piglets ingest IgA, the immune response of the intestinal mucosa is regulated to respond against intestinal pathogens (TIZARD, 2014).

Therefore, sows supplemented with a functional fiber capable of modulating the intestinal microbiota, produce responsive antibodies to specific pathogens. The maternal intestinal microbiota is recognized and encoded by specific antibodies present in the intestinal mucosa. IgA is transported into the bloodstream to the mammary gland, where it is complexed in milk. In turn, maternal IgA arrives the intestinal mucosa of piglets, where it will adhere to the intestinal mucosa to respond to pathogenic microorganisms (MACPHERSON et al., 2017; TIZARD, 2014).

The quality of the milk as well as its production by the sows are among the main factors associated with the performance of piglets in the maternity (TAN et al., 2018d). In addition, maternal health is also of great relevance in the performance results of piglets (ZHOU et al., 2018). That is way, the inclusion of functional foods, especially fiber of excellent quality, can ensure the health and well-being of the sows, and consequently maximize production by improving litter performance (TIAN et al, 2020).

However, in the present study, the use of dietary fibers in the gestation and lactation phases of sows were not able to improve any of the litter performance parameters. Yan et al. (2016) similar results when including high levels of resistant starch in the gestational diet of sows, where no change in piglet performance was observed during the lactation phase.

The findings of the present study contradict the results observed by Loisel et al. (2013), by including a mix of ingredients rich in insoluble fibers in the gestational diet of sows, observed a reduction in the percentage of pre-weaning mortality with the high inclusion of the fiber mix, thanks to the reduction in the duration of farrowing and the vitality of the piglets to suckle colostrum more quickly. Furthermore, the inclusion of the fiber mix was able to change the composition of colostrum and milk of the sows, with an increase in lipid and IgA levels. Shi et al. (2021) also observed higher average weight of piglets at weaning with the high dietary inclusion of NDF in the gestation phase. The authors attribute this result to the ability of fiber-rich diets to stimulate the increase in prolactin concentration during gestation, for the regulation of colostrum and milk production during the lactation phase.

The control treatment showed a tendentious result to the highest plasma urea level at 111 days of gestation. The other treatments showed lower levels plasmatic urea at 111 days of gestation. A similar result was observed by Shang et al. (2021), by including 20% of beet pulp in the diet of sows during gestation, where the authors founded a reduction in plasma urea levels at 110 days, compared to the control treatment. The authors also have no difference in serum triglyceride and total cholesterol levels.

In the large intestine, the protein that was not used along the digestive tract is transformed into ammonia, which can be used by microorganisms or absorbed and transported to the liver, where it is converted into urea (LEVRAT et al., 1993). Plasmatic urea can be considered an indicator of metabolic utilization of nitrogen in non-ruminant animals (OROK and BOWLAND, 1975). In the present study, the inclusion of S25F in the diet during gestation may have provided an improvement in nitrogen utilization. Another explanation would be the reduction in protein fermentation caused by dietary fiber, which caused a decrease in ammonia production (PIEPER et al., 2012).

In contrast the results of the present study, Weng et al. (2020) and their collaborators found changes in some blood parameters at 14 days of lactation in sows supplemented with different sources of dietary fiber. The authors reported higher plasma concentrations of total protein and triglycerides in sows that received the treatment with 20% rice husk. Higher levels of total proteins may be associated with immune responses (SZUBA-TRZNADEL et al., 2017).

The concentration of VFAs in feces represents approximately 5% of all VFAs production in the large intestine (MONTROYA, 2016). Although the percentage is small, the concentration of VFAs in feces is a very important evaluative parameter that is associated with the health and intestinal integrity of sows, as well as the fermentative quality of the digesta that arrives in the large intestine (TIAN et al 2020). Thus, it is believed that soluble fibers have a greater capacity to produce VFAs than insoluble fibers, which can favor the structuring of a microbial community beneficial to the host (JHA and BERROCOSO, 2015).

The fiber source can also change the proportion of VFA produced from the fermentation of dietary fiber. Acetic acid, for example, is produced in greater proportion in the fermentation of legumes, such as beet pulp and alfalfa flour. Cereals such as wheat

bran and corn bran produce a higher proportion of butyric acid as a fermentation product (LI et al., 2020a).

In the present study, at 111 days of gestation, acetic acid levels in feces were higher in the control treatment compared to the other treatments. This result may be associated with the composition of fiber present in experimental diets. The control treatment had soybean hull as a fiber source. The other treatments contained higher percentages of insoluble fiber. The studies by Li et al. (2019b) corroborate this affirmation, where higher levels of acetate were found in their results in the feces of sows that received diets with greater inclusion of soluble fiber, compared to diets with higher proportions of insoluble fiber.

Acetic acid is the most abundant microbial fermentation product, accounting for 90% of the total VFAs produced (JHA et al., 2019). This VFAs is the biggest contributor to the variation in microbial diversity (BEIRE, 2018). Acetic acid produced in the large intestine, when absorbed by the intestinal epithelium, is transported to the liver where it participates in glycolipid metabolism (FERNÁNDEZ et al., 2016). Furthermore, acetic acid is considered an anti-inflammatory metabolite and responsible for intestinal homeostasis (MARTENS, 2018; FUKUDA et al., 2011). Another important factor is that acetate serves as an energy substrate for butyric acid-producing bacteria. (LOOFT et al., 2014).

The levels of IgG and IgA in milk may also be related to the concentration of acetate produced by the intestinal microbiota through fiber-rich food. Kim et al. (2016a) observed that VFAs produced by the intestinal microbiota stimulate increased serum antibody levels in humans. Maternal antibodies are directed to the mammary glands, where they will be incorporated in the milk for the immunization of the offspring (TIZARD, 2014). These results can be compared with the findings of the present study, where higher levels of acetate in feces and IgG in the milk of sows that received the control diet were observed.

In the present study, the levels of butyric, propionic, valeric and isovaleric acid did not differ among treatments, both at 111 days of gestation and at 16 days of lactation in the sows. Leblois et al. (2018) also found no difference in the concentration of VFAs in the feces of sows at 106 days of gestation and in the second week of lactation, when supplemented with 33% resistant starch.

Although butyric acid was not relevant in this study, its importance is noteworthy, especially when it comes to the modulation of intestinal permeability (HUANG et al., 2015). Recent studies have reported on the production of butyrate by bacteria of the phylum Bacteroidetes ssp present in fecal samples from sows supplemented with soluble fibers (OCHOA-REPÁRAZ et al., 2010; TANOUE et al., 2016). The increased production of butyrate can stimulate the expansion of immune cells and the release of anti-inflammatory cytokines, such as IL-10 (CHENG et al., 2018). In addition to butyric acid, propionic acid is also considered one of the main metabolites produced by the intestinal microbiota.

The fecal pH values, between 7.0 and 8.0, measured in the present study may have interfered with the proliferation of bacteria that produce butyric acid. A pH range for the growth of bacteria producing butyric acid is around 5.5 (REICHARDT et al., 2018). The pH close to 8.0 increases the solubility of H₂S, releasing HS ions that inhibit the proliferation of bacteria producing butyric acid (RABA et al., 2021).

The isobutyric concentration was higher at 111 days in feces of sows supplemented with the control treatment, compared to the other treatments. The relevance and effects of isobutyric acid on swine intestinal health are poorly understood in the literature. In humans, it is known that the proportion of branched-chain fatty acids (BCFAs) produced by the intestinal microbiota is low (RIOS-COVIAN et al., 2020). These fatty acids are produced from the fermentation of amino acids by bacteria of the *Bacteroides* and *Clostridium* genus, and in greater intensity in the distal colon (SMITH and MACFARLANE, 1998; AGUIRRE et al., 2016).

Low concentrations of isobutyric acid and plasmatic urea at 111 days of gestation, referring to sows that received treatments with inclusion of functional fibers, can predict the efficiency of fibers in modulating protein metabolism. Studies carried by Aguirre et al. (2016) suggest that the production of isobutyric occurs with the products of protein fermentation in the colon, such as ammonia, for example. Ammonia, when produced in excess, can damage the intestinal environment (GILBERT et al., 2018). In addition, the ammonia produced in large amounts can be toxic to sows in the last third of gestation (HERRING et al., 2018).

In a study conducted by Rios-Covian et al. (2020), a negative correlation was reported between fiber intake, especially insoluble fibers, and isobutyric levels in adult

humans. The same can be observed in the present study, where isobutyric levels were lower with the inclusion of S25F and IF. The arabinogalactan present in S25F is a polysaccharide able of reducing the synthesis of isobutyric in the colon, through the reduction of protein fermentation that generates metabolites harmful to health, as previously mentioned (CHEN et al., 2021c).

The intense metabolic activity during the lactational phase of the sows may be a possible explanation for why the results of VFAs at 16 days of lactation were not expressive. It can be inferred that the use of nutrients during the lactation phase was efficient for milk production and body maintenance. sows, as previously mentioned, did not suffer significant body losses and milk production was within normal limits. Therefore, it can be affirmed that the VFAs produced during lactation were used by the metabolism of sows.

Fecal pH can be considered an important parameter related to the condition of the intestinal microenvironment, essential for homeostasis and favorable growth of beneficial microorganisms. The pH of swine feces under healthy conditions will be alkaline and may vary according to diet, age, and physiological condition (SIBA et al., 1996). When it comes to diet, the inclusion of fibers such as inulin can favor the synthesis of VFAs, ending up in acidification of the large intestine (HANSEN et al., 2011b).

Excessive acidification of the digesta in the large intestine can cause irritation and inflammation in the mucosa, causing an increase in intestinal permeability and, consequently, an increase in osmotic pressure (BROWN et al., 2007b). The acidification of the intestinal lumen can occur through the high production of VFA or by the excess of acid metabolites in response to dysbiosis (GARBOSSA et al., 2019). In the present study, the inclusion of functional fiber in the diet of sows did not change fecal pH, with all treatments remaining between 7.0 and 7.5.

In the peripartum, constipation can be one of the main aggravating factors that can harm the health of sows and piglets during the farrowing. The use of good quality fibers such as S25F and IF fibers have the positive point of increasing the passage rate of the digesta, favoring the faster passage of piglets through the uterus without prolonging the duration of the farrowing. In the present study, stool dry matter was measured to evaluate the effects of inclusion of functional fibers on the percentage of stool moisture. However,

the inclusion of functional fibers did not change the percentage of humid in the feces of the sows, both in peripartum and lactation.

The inclusion of fiber in the diet during gestation, regardless of its physicochemical composition, induces water retention in feces, relieving the level of constipation, especially in the peripartum (FEYERA et al., 2017; SHANG et al., 2021). This affirmation was reported by Tan (2015a) and his collaborators, when evaluating the fecal score throughout the lactational phase of sows that received a diet including 2.2% Konjac flour during gestation. However, the inclusion of fiber sources with a higher proportion of insoluble components stimulates the secretion of water and mucosa in the intestinal lumen, which alters the animals' fecal score (MCRORIE et al., 2015).

Nutrition is one of the main factors that can alter the gastrointestinal tract microbiota. As well as nutrition, the composition of ingredients can modulate the microbiota in benefit of the individual's health, by stimulating the production of essential metabolic components (MARKOWIAK and ŚLIŻEWSKA et al., 2018). Functional fibers can be considered as ingredients able to modulate the intestinal microbiota, by stimulating the multiplication of beneficial microorganisms to maintain intestinal eubiosis (BERROSO et al., 2015).

But first, knowing the composition and co-evolution of the intestinal microbiota and host is extremely important for animal health (ELSON and ALEXANDER, 2015; WOO and ALENGHAT, 2017). Thus, the microbial composition of the gastrointestinal tract may be closely linked to the reproductive phase of animals, with notable changes during gestation and lactation (CHENG et al., 2018; LIU et al., 2019).

In the last third of gestation, the composition of the microbiota of sows is mainly composed by the phyla Firmicutes and Bacteroidetes (CHEN et al., 2017a). These findings corroborate the results of the present study. In all treatments, the relative abundance of these phyla was higher compared to the other phyla.

In the present study, the Euryarchaeota phylum was the only one with relative abundance altered with the inclusion of dietary functional fiber. The relative abundance of the Euryarchaeota phylum was greater at 111 days of gestation and 16 days of lactation, with the inclusion of S25F fiber. The treatment with IF inclusion showed the lowest relative abundance of the Euryarchaeota phylum.

The Euryarchaeota phylum is mainly composed of methanogenic archaea, associated with the fermentation of non-digestible polysaccharides in the colon to reduce carbon dioxide into methane (TRIANAFYLLOU et al., 2014). In humans, increased methane production can be associated with enteric diseases such as intestinal inflammation and constipation and metabolic disorders such as obesity (KUNKEL et al., 2011; PIMENTEL et al., 2003).

At 111 days of gestation, the inclusion of functional fibers reflected the relative abundance of bacteria of the genera *Clostridium IV* and *Terrisporobacter*, belonging to the phylum Firmicutes, and *Methanobrevibacter*, belonging to the phylum Euryarchaeota (the only identified phylum that belongs to the Archaea domain). The relative abundance of the genus *Clostridium IV* was higher in the feces of sows fed diet with S25F inclusion. In general, the clostridia are acidogenic bacteria capable of producing acetic and butyric acid (SHIDA, 2008). Bacteria belonging to the genus *Clostridium cluster IV* are recognized as butyric acid-producing bacteria and are generally associated with a microbiota considered to be healthy (FUENTES et al., 2014; ALOU et al., 2017). To maintain intestinal fermentation, it is crucial that there is a balance between hydrogenotrophic and hydrogenogenic bacteria. Acetogenic bacteria can convert butyric and propionic acid into acetic acid, generating substrate for methanogenic bacteria to produce methane or acetic acid (SMITH et al., 2018).

Archaea of the genus *Methanobrevibacter* characterized as methanogenic and hydrogenotrophic bacteria, in addition to controlling the concentration of acetic acid, also modulate the pH of the environment, removing hydrogen from the system (SHIDA, 2008). This genus is found naturally in the gut of humans and monogastric animals and plays an important role in the efficient digestion of polysaccharides by metabolizing microbial fermentation products (LUO et al., 2012; ARMOUGOM et al., 2009). This genus was also observed in greater abundance in the feces of sows that received the diet with S25F inclusion. There was a better use of fiber present in the diet by favoring the growth of *Methanobrevibacter* archaea.

S25F contains 25% of hemicellulose arabinogalactan, a source of polysaccharide also used by *Methanobrevibacter* for methane synthesis (LI et al., 2018c). By making the source of hemicellulose available in the intestinal lumen, proliferation of

Methanobrevibacter may have been favored, due to their metabolic dependence on other metabolites (FEDERICI et al., 2015).

Archaea of the genus *Methanobrevibacter* were also observed in greater abundance at 16 days of lactation in sows that received the inclusion of S25F. However, the abundance values found at 16 days were lower than at 111 days of gestation. The reduction in the *Methanobrevibacter* population may be associated with the intense metabolism found in the lactational peak of sows. The greater the milk production, the greater the maternal energy demand (STRATHE et al., 2017). Thus, the absorption of VFAs, an energy source for microorganisms, will be scarcer, forcing a reduction in the proliferation of microorganisms dependent on the consumption of these metabolites (MCCORMACK et al., 2017; WEISS et al., 2017).

Furthermore, sows in the lactation period are in constant metabolic activity, which can generate a basal inflammatory response, due to the need for milk production. Metabolic inflammation in sows with high milk production may be associated with lower intestinal microbial richness (SHAO et al., 2020; LE CHATELIER et al., 2013). As an example, the relative abundance of the phyla Bacteroidetes and Firmicutes is associated with the host's energy metabolism, due to the high energy demand for lactogenesis (LEY et al., 2006; KOMAROFF, 2017).

In studies with animal models, the genus *Methanobrevibacter* has been correlated with obesity, and in humans, greater abundances were observed in individuals with high body mass index (SINGER-ENGLAR et al., 2018). Considering that the maternal gut microbiota can be transferred to piglets through feces, studies have reported that archaea of the genus *Methanobrevibacter* may be associated with overweight in children (MBAKWA et al., 2015; MATHUR et al., 2013).

The genus *Terrisporobacter*, previously classified as *Clostridium sp*, has acetic acid from simple carbohydrates as its main product (LUND et al., 2015, GERRITSEN et al., 2014). Some reports in humans revealed bacteria of the genus *Terrisporobacter* as opportunistic pathogens (CHENG et al., 2016). In the present study, bacteria of the genus *Terrisporobacter* were found to be less prevalent in the feces of sows that received a diet including S25F at 111 days of gestation. This result reveals a positive outlook in favor of the S25F fiber, by modulating the microbiota to lower proportions of pathogenic bacteria present in the intestinal microbiota.

At 16 days of lactation, a greater relative abundance of the genera *Clostridium IV* and *Clostridium cluster XIVa* was observed in the feces of the sows that received the control treatment. The decrease in the percentage of S25F and IF inclusion, from 2.5% to 1% in the lactation diet, may be related to the reduction in the relative abundance of microorganisms. The reduction in the abundance of these microorganisms may be associated with the need for higher levels of inclusion of functional fibers during the lactation phase. For example, high fiber diets can increase the population of cellulolytic bacteria (CAO et al., 2016). Cao et al. (2016) reported the effects of low-fiber diets and their composition on increasing the population of *Methanobrevibacter* in gilt feces.

The effects of S25F inclusion on the relative abundance of *Methanobrevibacter* archaea can be observed at 16 days of lactation, where sows that consumed the S25F diet had a higher prevalence of this genus. This result may be related to the physicochemical properties of S25F fiber. The difference among treatments is in the higher concentration of arabinogalactans present in S25F.

5. CONCLUSION

The inclusion of eubiotic fiber in the diet during gestation and lactation do not change the performance of the sows and their respective litters. However, plasma urea levels at 111 days are lower compared to the control treatment, that is, it improves the protein digestibility of the diet. Fiber inclusion also reduces isobutyric acid synthesis, providing better intestinal health.

In addition, eubiotic fiber can alter the intestinal microbiota of sows, stimulating the growth of bacteria that improves the use of fibers and nutrients present in the diet and reduces the population of pathogenic bacteria. The sows that receive a diet with the inclusion of eubiotic lignocellulose can transfer a higher percentage of IgA to the milk, ensuring the piglets a greater intestinal immunological support.

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