



**PIG HEALTH:
MORE THAN JUST
INTESTINAL ACTION**



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SUMMARY

THE IMPORTANCE OF INTESTINAL MICROBIOTA IN PIGS

Microbiota modulation: a major asset for pig health

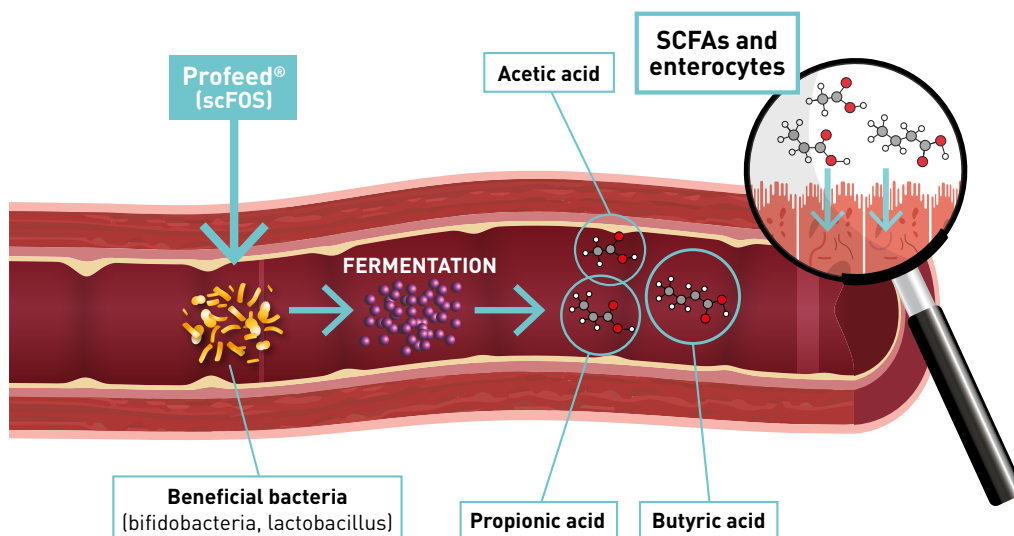
The post-weaning period is a critical period accompanied by nutritional social and environmental stress. The diet passes from a highly digestible liquid diet (i.e., milk) to a solid, more complex, and less digestible diet. The change is also marked by a switch from fat towards carbohydrates as main source of digestible energy. During this period, faecal diarrhea scores not only reflect ingredient quality and starter feed formulation, but also microbiota balance. Indeed, dietary modifications may result in disrupted gut microbiota composition and intestinal inflammation, which can lead to the growth of enteric pathogens with consequences on post-weaning diarrhea.

Mammalian intestine is estimated to contain approximately 10^{10} to 10^{14} microorganisms, about 10 times more than the number of cells composing the host body!

Profeed® is composed of short-chain fructo-oligosaccharides (scFOS), known to be prebiotic fibres, and helps to have well-balanced microbiota.

The large size of microbiota has been known for a long time. This mutually interacting system composed of the host cells and the resident microbes is called intestinal **microbiome**. Intestinal microbes play a crucial role in host health. They act as a defending barrier against invading pathogens, aid digestion and energy harvest from the diet, provide nutritional support for enterocytes, and stimulate the development of the local immune system.

Figure 1 : Production of short-chain fatty acids (SCFA) through Profeed® fermentation by beneficial bacteria of the gut microbiota



Profeed® allows modulating pig intestinal microbiota thanks to:

- **Selective growth of beneficial bacteria and exclusive competition** with pathogens for a well-balanced microbiota
- **Greater short-chain fatty acids (SCFAs) production.** Produced by fermentation, SCFAs are the preferred fuel source of enterocytes, playing a crucial role in digestive process and protection of the intestinal barrier.

ISAPP*
definition 2010

"A dietary prebiotic is a selectively fermented ingredient resulting in specific changes in the composition and/or activity of gastrointestinal microbiota, thus conferring benefit(s) upon host health"

*International scientific association for prebiotics and probiotics

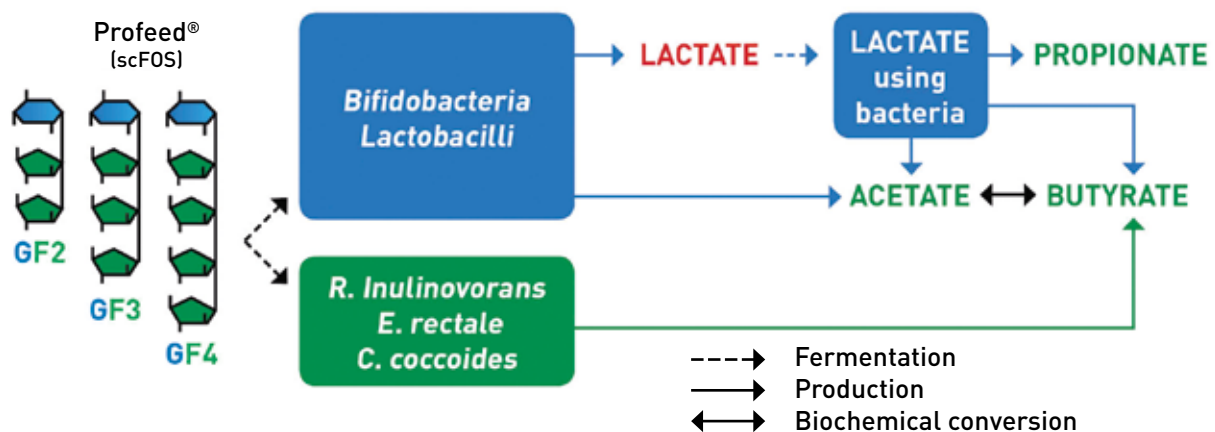
Profeed® promotes selective growth and exclusive competition...

In the eubiotic status, the intestinal microbiota is characterized by a preponderance of beneficial species [1]. By its selective action, Profeed® allows promoting growth of beneficial bacteria; while it is not used for the growth of potentially pathogenic bacteria. In addition, the growth of potentially beneficial bacteria induces competitive exclusion resulting in lower presence of potentially pathogenic bacteria.

Intestinal health depends on the balance of bacterial species, also called eubiosis. Through its selective action, Profeed® is in favour of the status of eubiosis.

In a study in young piglets (7 days of age) fed with milk replacer with or without a daily dose of 1 g Profeed®, count of *E. coli* significantly decreased (-1 log at 10 days) while Bifidobacteria count increased with Profeed® supplementation compared to the non-supplemented group. When increasing, Bifidobacteria may exert competitive exclusion leading to decreased *E. coli* populations [2].

Figure 2 : Mechanism explaining the increase of SCFA and the transient increase of lactate with Profeed®



By its selective fermentation, Profeed® allows increasing short-chain fatty acids production.

Regular supplementation with Profeed® allows increasing SCFA concentration and faecal dry matter content in piglets [3]. SCFAs, and especially butyrate, play a crucial role in enterocyte growth, and are used as main fuel for immune cells. Interestingly, supplementation with Profeed® increases butyrate concentration and bacteria population such as *Megasphaera elsdenii*, lactate-using bacteria in piglets aged 40 days [4]. A positive correlation between butyrate, the number of crypt epithelial cells, and crypt depth is shown, underlining the importance of butyrate for gut mucosa.

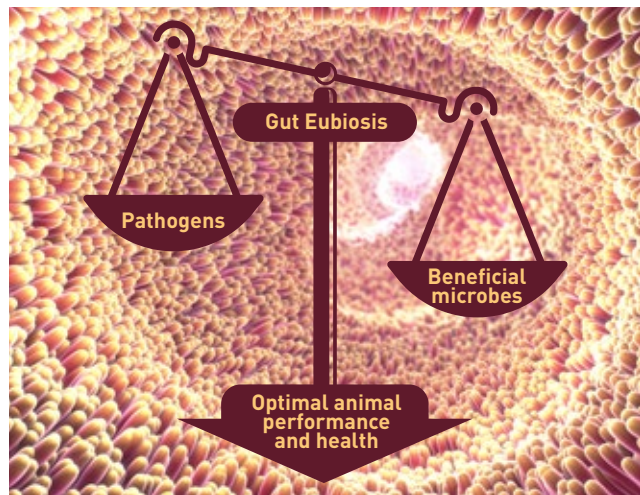


INTESTINAL MICROBIOTA IS THE HUB OF OVERALL HEALTH

There are numerous interactions between microbiota and its host, underlining the importance of an eubiosis status for the host health.

Modulation of intestinal microbial populations has important functional consequences as it strongly interacts with the host digestive, immune and metabolic functions.

This modulation can be of importance in the context of decreasing antibiotic use. Indeed, microbiota modulation can be beneficial for gut barrier function, the immune system and productive performance.



Beyond antibiotherapy, there are only 3 ways to manipulate microbiota:

Prebiotics

«A selectively fermented ingredient that results in specific changes, in the composition and/or activity of the gastrointestinal microbiota, thus conferring benefit(s) upon host health».

ISAPP 2008/IFIS Functional Foods Bulletin 2011

Probiotics

«Live microorganisms which when administered in adequate amounts confer a health benefits on the host».

FAO/WHO 2001

Faecal transplantation

Faecal transplantation (or bacteriotherapy) is the transfer of stool from a healthy donor into the gastrointestinal tract for the purpose to treat recurrent diseases.

Synbiotic:

← Combination of prebiotics and probiotics →

Prebiotics are the most efficient and easiest way to well-balance microbiota in pigs. They can be given in feed daily. Beyond the effect on microbiota composition and activity, other beneficial effects are highlighted with Profeed®.

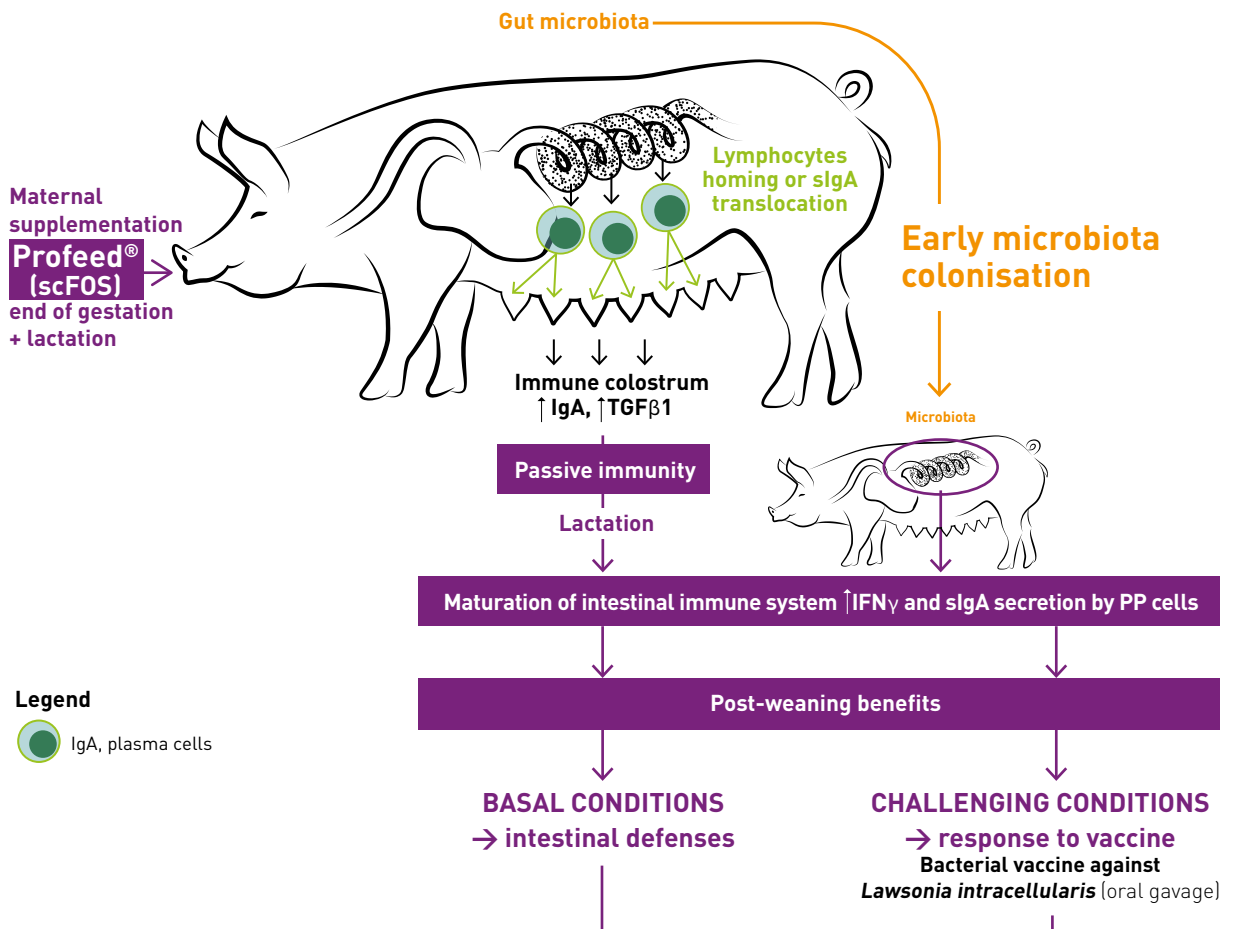
Beyond modulating pig intestinal microbiota, a daily supplementation with Profeed® also supports:

- › Immune system maturation and functionality in neonatal suckling piglets
- › Immune response to different types of challenges in suckling and weaned piglets
- › Reproductive performance and health of sows
- › Growth performance of weaned piglets

Profeed® beyond intestinal microbiota modulation: immune transfer from mothers to their piglets

Studies conducted in sows and their neonatal piglets reported an effect of Profeed® on immune transfer from mothers to young, on maturation of immune and gut systems, and on immune response after intestinal challenges in weaned piglets.

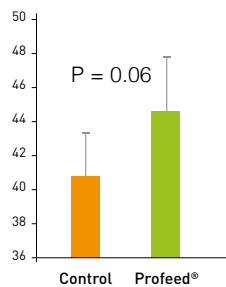
Figure 3 : Effects of maternal Profeed® supplementation on the intestinal immune system of piglets. Supplementation : 0.33% Profeed® during the last 4 weeks of gestation and 0.15% during the entire lactation period [5].



Legend
 IgA, plasma cells

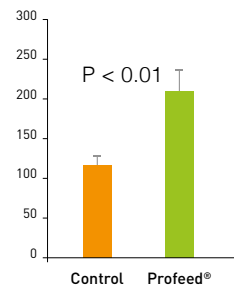
These studies demonstrated the effectiveness of indirect nutritional intervention, i.e. maternal supplementation with Profeed®, to improve offspring vaccine response towards intestinal bacterial challenge unveiled by increased vaccine-specific IgA levels in serum and ileal mucosa of weaned pigs.

Number of goblet cells per crypt in caecum



Maternal Profeed® supplementation tends to increase the number of goblet cells in caecum (P = 0.06). Goblet cells are specialised epithelial cells secreting mucin glycoproteins involved in the maintenance of intestinal mucosal surface integrity.

Specific IgA response to *Lawsonia intracellularis* vaccination in serum (arbitrary unit)



Maternal Profeed® supplementation results in faster gut immune system maturation and allows significantly improved immune response after vaccination against *Lawsonia intracellularis*, an enteric pathogenic bacteria in weaned piglets (56 days of age; [6]).

Profeed[®], beyond intestinal microbiota modulation: positive effects of direct supplementation on gut immunity in suckling piglets

Studies conducted in suckling piglets directly supplemented with Profeed[®] reported an effect on gut barrier function and on the immune system, resulting in :

- decreasing mortality in the post-natal period
- improving the inflammatory and immune status

A study conducted in newborn piglets from 17 litters (sow parity between 2-8, ≥14 piglets born alive) randomly allocated within blocks of birth weight to 3 treatments (daily drenching with pipet and a flexible 7 cm tube of 2 mL placebo solution (T1) or 1 g Profeed[®] solution for 7 days after birth (T2) or until weaning (T3, 21 days; [7]) showed Profeed[®] allows modulating inflammatory and immune factors and improving survival.

Figure 4 : Inflammatory markers in intestinal mucosa and faeces

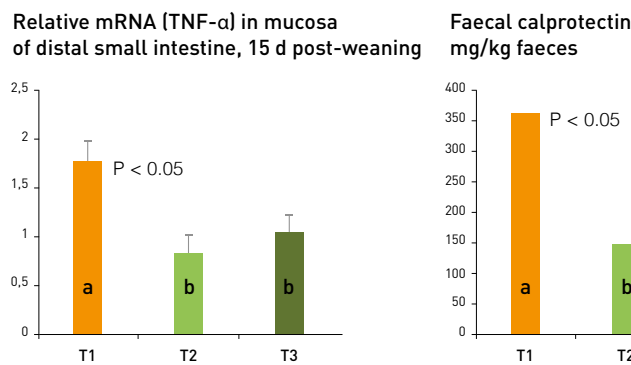


Figure 5 : IgG concentration in serum

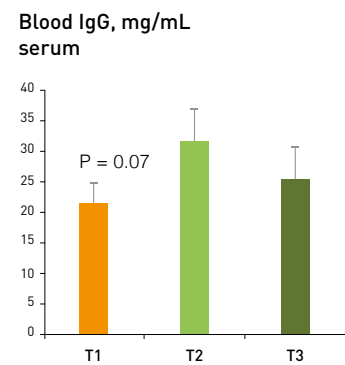
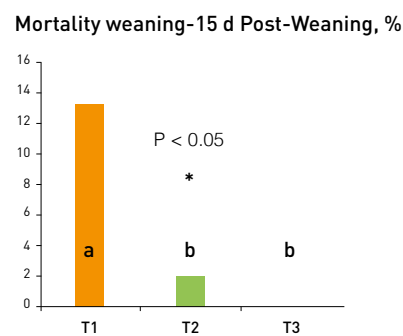


Figure 6 : Mortality rate during lactation period



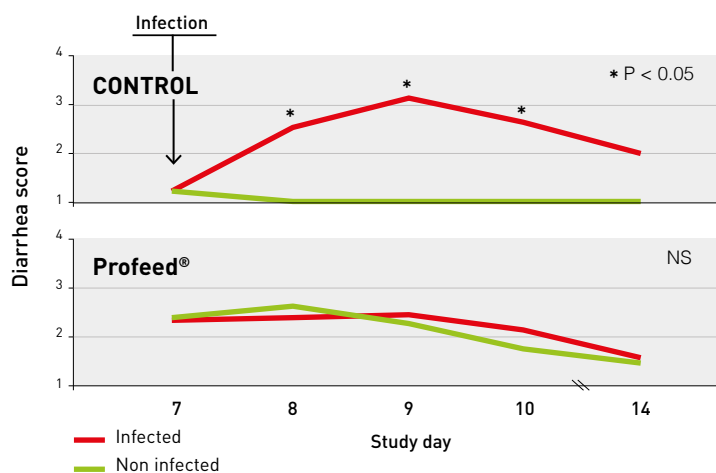
Faecal calprotectin and TNF-α are used as inflammatory stress markers. These 2 parameters decrease with Profeed[®] supplementation, suggesting Profeed[®] allows modulating inflammatory state. Furthermore, blood immunoglobulin G (IgG) content tends to be higher with Profeed[®].

The effects observed on inflammatory and immune status are accompanied by a significant decrease in the mortality rate [7].

Profeed[®] allows reducing morbidity following *S. typhimurium* or *E. coli* infection in neonatal piglets

Two studies conducted in neonatal piglets fed with milk replacers with or without Profeed[®] [2; 8] and challenged with either *Salmonella typhimurium* [8] or *E. coli* K88 [2] showed Profeed[®] significantly reduces morbidity of infected piglets, while improving diarrhea scores.

Figure 7 : Diarrhea score in non-infected and infected piglets

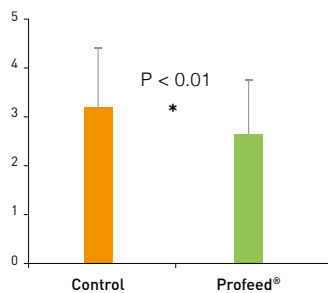


Profeed[®], beyond intestinal microbiota modulation: benefits for sows

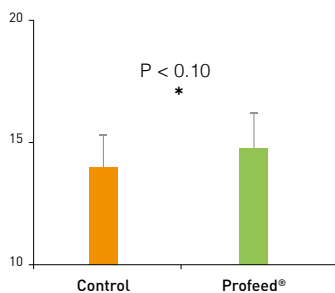
A daily supplementation with Profeed[®] allows reducing farrowing duration and decreasing fat mobilisation in sows during lactation. These results could reflect a better physiological status of sows.

Figure 8 : Effect of Profeed[®] in sow's diet on farrowing duration and backfat thickness at weaning

Farrowing duration, h



Backfat thickness at weaning, mm

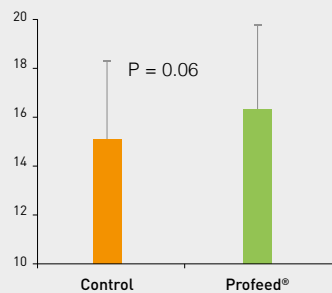


In a commercial farm, for sows receiving 0.33% Profeed[®] the last 5 days of gestation and 0.15% Profeed[®] during the entire lactation period, the farrowing duration is shorter and backfat is thicker at weaning [5; 6].

Furthermore, a tendency to increase prolificity was also observed during the reproductive cycle following supplementation with Profeed[®][9].

A daily supplementation with Profeed[®] allows improving prolificity of the reproductive cycle.

Figure 9 : Number of piglets born during the next reproductive cycle



Supplementation with 0.33% Profeed[®] in late gestation and 0.15% during lactation results in benefits for sows and piglets.



Profeed[®], beyond intestinal microbiota modulation: performance in weaned, growing and fattening pigs

Supplementation of weaned piglets with Profeed[®] between 0.1% and 0.4% allows improving average daily gain without modifying daily feed intake.

A meta-analysis has been performed on 7 studies (22 data) to establish law of response of growth performance to supplementation with Profeed[®] in weaned piglets.

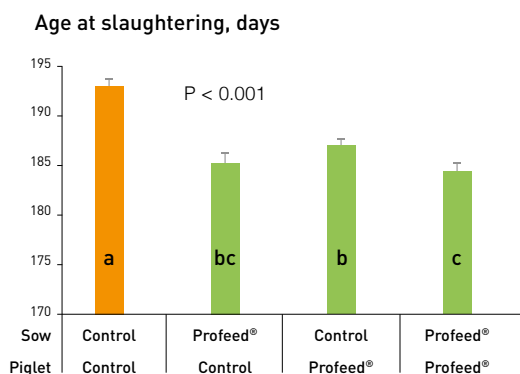
| Characteristics of the 7 studies used to perform the meta-analysis | | | | | | | |
|--|------------|-------------|-------------------|-------------------------|--------|---------|------|
| N = 22 | Profeed, % | Duration, d | Age of piglets, d | Initial body weight, kg | ADG, g | ADFI, g | FCR |
| Mean | 0.19 | 33.9 | 20.3 | 5.99 | 371 | 573 | 1.55 |
| Minimum | 0 | 21.0 | 15.5 | 4.85 | 249 | 377 | 1.39 |
| Maximum | 0.71 | 49.0 | 33.0 | 7.87 | 463 | 717 | 1.90 |
| Standard deviation | 0.21 | 10.2 | 5.5 | 0.88 | 61.6 | 102.5 | 0.12 |

Figure 10 : Average daily weight gain according to the dose of Profeed[®] supplementation



Perinatal supplementation with Profeed[®] results in a carry-over effect until slaughtering, leading to higher profits for breeders.

Figure 11 : Effect of maternal and/or post-weaning Profeed[®] supplementation on the age of commercial slaughtering



Perinatal supplementation with Profeed[®] of sows in late gestation (0.33%), for the whole lactation period (0.15%), and of piglets during the starter phase (0.15%) resulted in a strong carry-over effect on growth performance, as piglets more rapidly reached the target body weight of 120 kg when they received Profeed[®] or when their mother received it [9].

These results suggest a long-term effect of early supplementation with Profeed[®].

Supplementation with 0.1 - 0.15% Profeed[®] at early stage allows improving growth performance of pigs.

Profeed[®], beyond intestinal microbiota modulation: what about reducing antibiotic use?

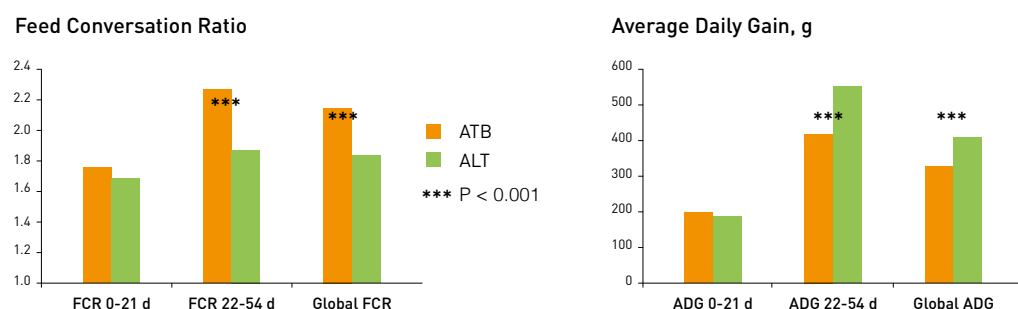
The development of alternatives to the use of antibiotics has become a major concern in recent years. This research can mobilise many areas: genetic selection, vaccination, and nutrition. These alternatives, and especially nutrition, are essential when focused on periods of high stress for animals, such as weaning.

Thanks to its action on gut microbiota and its immunomodulatory effects, Profeed[®] is of strong interest to reduce antibiotic use.

A combination of protected organic acids and essential oils (P(OA+EO)) associated with Profeed[®] and protected zinc oxide (P(ZnO)) is used as an alternative to antibiotics or high zinc oxide doses in weaned piglets [10]. On an experimental farm, 480 piglets aged 19 days were randomly allocated to either of the following feeding programs: **1)** ATB, consisting in a pre-starter diet supplemented with 120 mg colistin/kg feed, 300 mg amoxicillin/kg feed, and 2.400 mg ZnO/kg feed, and then a starter diet supplemented with 1 600 mg ZnO/kg feed; or **2)** ALT, consisting in an antibiotic-free (ABF) pre-starter feed supplemented with 1 g Profeed[®]/kg feed, 2.5 g P(OA+EO)/kg feed, and 0.25 g P(ZnO)/kg feed, followed by an ABF starter feed supplemented with 1.5 g P(OA+EO)/kg feed, and 0.25 g P(ZnO)/kg feed. Both ATB and ALT diets were distributed for 21 and 33 days for the pre-starter and starter diets, respectively.

| | ATB diet | ALT diet | P-value diet |
|-------------------------|----------|----------|--------------|
| BW at weaning, kg | 6.7 | 6.7 | NS |
| BW at 21 days, kg | 10.7 | 10.6 | NS |
| BW at 54 days, kg | 24.2 | 28.6 | < 0.001 |
| Diarrhoea score 0-21 d | 0 | 0.7 | Not tested |
| Diarrhoea score 22-54 d | 0 | 0 | Not tested |

Figure 12 : Performance of piglets during lactation and starter phase after weaning



There was no difference on growth performance during the pre-starter phase. The diarrhoea score was higher in ALT group than in ATB group, suggesting piglets were more challenged. However, average daily gain (ADG) and feed efficiency (FCR) were significantly improved during the starter phase, leading to better overall FCR and ADG. Thus, in spite of higher microbial challenges at the beginning, piglets receiving an alternative diet to antibiotics showed better growth performance.

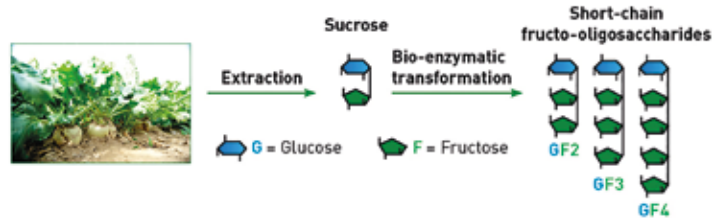
It can be concluded that including 0.1% Profeed[®] in combination with other ingredients to reduce antibiotic use and high level of zinc oxide can support good growth performance of weaned piglets.

WHY IS PROFEED® THE IDEAL PREBIOTIC FOR PIGS?

There are several prebiotics corresponding to the ISAPP definition BUT they do not have the same benefits depending on the species.

Short-chain fructo-oligosaccharides (scFOS) are natural ingredients that are found in small amounts in some fruit, vegetables or plants such as bananas, wheat, onions, and asparagus.

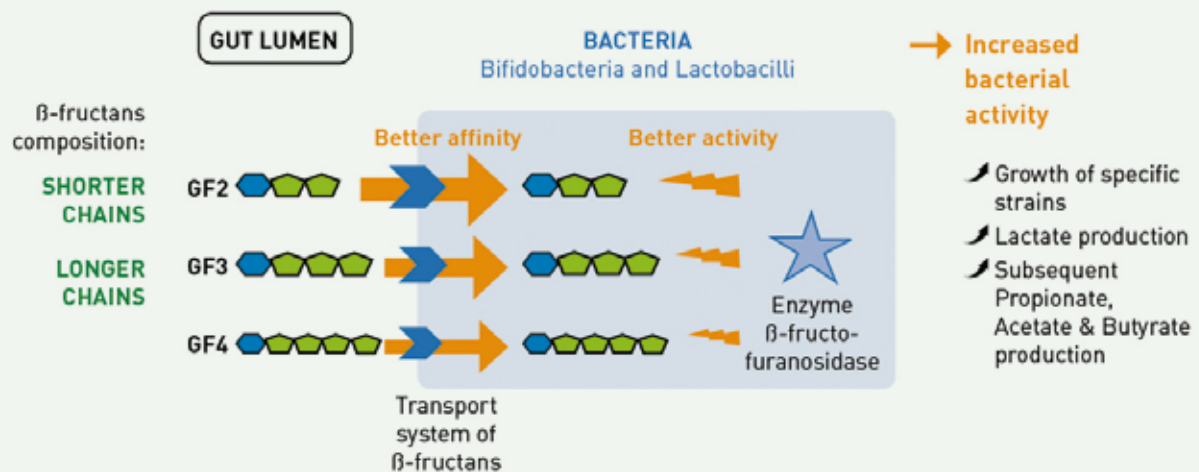
Profeed® is made up of scFOS obtained from beet sugar through a biotransformation leading to the formation of **three components, GF2, GF3 and GF4, with constant and guaranteed ratios.**



The very precise composition of Profeed® obtained by biosynthesis allows having predictable effects.

Profeed® is selectively fermented by some strains of Lactobacilli and Bifidobacteria. Several studies have explored mechanisms for use of Profeed® by Lactobacilli and Bifidobacteria to provide greater understanding of the selectivity shown by specific strains [11-13]. They demonstrated that the strong affinity of β -fructofuranosidase and transport mechanism of Lactobacilli and Bifidobacteria strains for short-chains of Glucose-Fructose explain the selective fermentation of scFOS compared to molecules with longer chains.

Figure 13 : Mechanism explaining selective growth of Bifidobacteria and some Lactobacilli with Profeed®



PROFEED®, A PERFECT PREBIOTIC FOR PIGS

- › Profeed® exerts a prebiotic effect at low dose, from 0.1%
- › Profeed® will be preferentially fermented by different bacteria strains
- › Profeed® fermentation occur in ileum and colon before reaching the distal part of the colon.
- › Profeed® benefits can be expected sooner in the digestive process, with no risk of possible negative consequences (like soft stools).

Your claims using Profeed® supported by our scientific dossiers

Many studies have validated and quantified the benefits of Profeed® and we continue to conduct research programs with Profeed®, to develop scientific substantiation.

We have defined and implemented a reliable strict approach to substantiate these feed claims by:

- exhaustively reviewing the bibliography,
- carrying out scientific studies in collaboration with recognized research institutes and universities,
- applying the latest evaluation techniques to ensure that relation between the feed material and the claimed effect is properly established and science-based.

Today, farmers are looking for more than just feed. They expect the feed they buy to have an added value on their animal's health and performance... Profeed® can help you to make your products healthier.

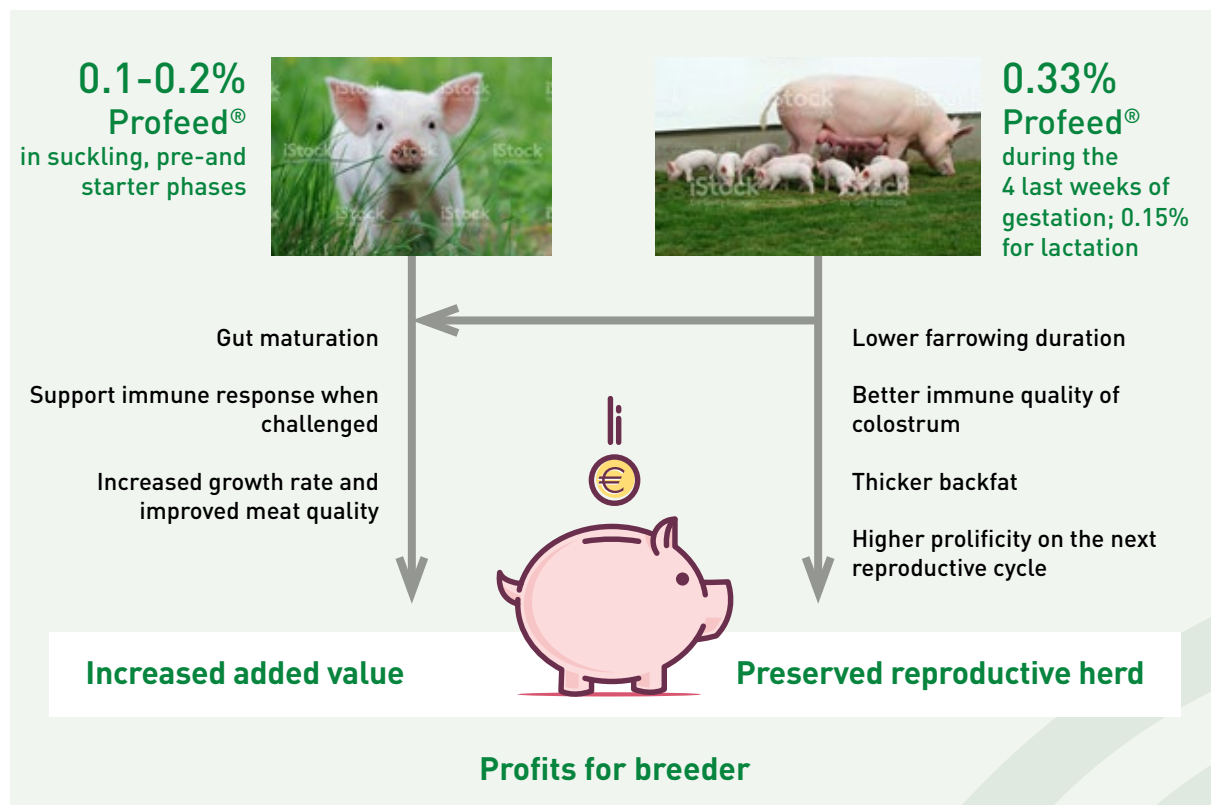
Our approach is in accordance with the European regulation 767/2009, which makes recommendations to establish health claims for feed.

- Do "fructo-oligosaccharides" appear in the composition list of your labelling?
- Is your feed material well produced from beet sugar through enzymatic process?



If you answer positively to the 2 questions above, you will be able to benefit from Tereos expertise, supported by scientific dossiers on how to better use claims for your feed products.

We support you for optimal use of Profeed® in your products and display comprehensive benefits for animals.



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