Effect of Certain Phytobiotics on the Digestive Microbiota of Broilers (Gallus gallus)

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Abstract: ***The use of growth-promoting antibiotics (GPAs) has been banned in livestock feed due to the increased risk of antibiotic resistance, allergy and to toxicity for both humans and animals. Our aim was to study the microbiological effect of phytobiotics (EO) on the digestive microbiota of broilers, turkeys and laying quails. The phytobiotics studied were fennel (Foeniculumvulgare), flax (Linum usitatissimum), rosemary (Rosmarinus officinalis) and thyme (Thymus vulgaris). The results showed that in broiler chickens, the phytobiotics fennel, flax, thyme and rosemary had a positive impact on reducing the digestive microbial population, in particular pathogens. These results open up a promising avenue of research into additives with bioactive substances as substitutes for risky conventional products, both for improving the microbial ecosystems of the poultry digestive tract and for improving the zootechnical performance of farm animals.***

Keywords: Phytobiotics, Feed additives, Microbiological effect and Broilers

1. Introduction

The use of antibiotics in animal nutrition as growth promoters has undoubtedly been beneficial for the zootechnical improvement of performance parameters and disease prevention. However, biosecurity threats to human and animal health resulting from increasing pathogen resistance to antibiotics and the accumulation of antbiotic residues in animal products.

The value of the raw materials made available to farm animals depends on the quality and extent of the host animal's microbial load, particularly in its digestive tract and environment. Unlike ruminants, poultry do not have a natural bacterial flora capable of degrading all nutrients. Poultry have limited resistance and immunity to infection by potentially pathogenic microorganisms. For this reason, the use of antibiotics as growth promoters and inhibitors of pathogenic bacteria has been recommended to improve production and health performance (Mckellar, 2004).

However, researchers have shown that the repeated use of antibiotics encourages the selection and proliferation of resistant micro-organisms. Especially since antibiotic feed additives for animals are of ten identical or similar to the active ingredients used in human therapy. In any case, their use is likely to lead to the emergence of bacterial resistances that can be disseminated in the environment or the food chain (Hillman, 2006; Diarra et al., 2007), which could represent a risk to human health. Some of them could promote the development of pathogenic bacteria through the reduction of commensal microbiota (Asakura et al., 2001). They can also be found in small quantities in animal meat (Kan and Meijer, 2007; Wise et al., 2007), or in large quantities in animal waste. On the other hand, the treatment and prevention of infections is likely to become increasingly difficult in both animals and humans, especially as the discovery of new antibiotic active ingredients becomes rarer (Aarestrup et al., 2008; Binh et al., 2008).

Antibiotic resistance and pharmaceutical residues are a major public health problem. This is no longer possible, however, following the findings of the WHO, OIE and FAO (Agisar, 2004). Since 2006, the European Union has systematically banned the use of CFCs in animal feed, whereas the ban in Morocco only began in 2013. Stopping the use of CFCs in poultry has, however, had negative consequences for poultry farming, leading to a reduction in growth performance, the development of diseases such as necroticenteritis and dysbiosis, and an increase in the use of therapeutic antibiotics for preventive purposes (Huyghebaert et al., 2011).

It is in this context that aromatic and medicinal plants (AMPs) and their extracts, also known as phytobiotics or phytogenics, have been the focus of research to assess the value of incorporating them into animal feed as non-antibiotic growth promoters, such as probiotics, prebiotics and symbiotics. The latter already have their place in animal nutrition.

2. Material and Methods

2.1. Location and study period

Poultry samples were taken at the Station Pédagogique Avicole of the Institut Royal des Techniciens Spécialisés en Elevage de Fouarat (IRTSEF), Kénitra-Maroc, with particular emphasis on ileal and caecal intestinal contents. The samples taken were submitted for microbiological analysis at the Department of Microbiology and Food Hygiene of the National Institute of Hygiene, Rabat, 2016.The animals recruited in this investigation are day-old chicks belonging to the Gallus gallus species, These animals were chosen as they represent the main cash poultry species as an animal model to better approximate the professional reality of the poultry sector and in order to understand the mechanisms related to the microbiological effects following the addition of essential oils in their diet

2.2. Feed

Animal feed is based on cereals and their by-products, oilseed cakes, fishmeal and premixes. Phytobiotics are used in the form of essential oils.

2.3. Experimental protocol

A total of 150-day-old chicks of Ross strain were divided into 6 groups of 25 individuals each. The animals were housed under the same conditions as Experiment I. Additives consisted of antibiotic (Oxytetracycline) and EO of fennel (Foeniculum vulgare), linseed (Linum usitatissimum), rosemary (Rosmarinus officinalis), and thyme (Thymus vulgaris). The animals were treated with 0.5 g/kg of the various test products. The choice of the dose in question was based on the results obtained from studies carried out by our team on turkeys (Ould Sidi Moctar *et al.*, 2015).

3. Results

3.1. Bacteriological effect on the cecum

The effect of different treatments on the bacterial community in broiler caeca showed variability in terms of bacterial numbers (log10UFC/g content). The results relating to the effects of these HE on the level of caecal content concentration by the species studied are reported in Figures 1, 2 and 3. Comparison of the means of the different species was carried out using the ANOVA test in SAS software, enabling us to distinguish between groups unaffected by the different treatments $p>0.05$ (fig. 1) and those showing significant differences $p<0.05$ (fig. 2 and 3). At first glance, these results show variability depending on the treatment involved for the same site.

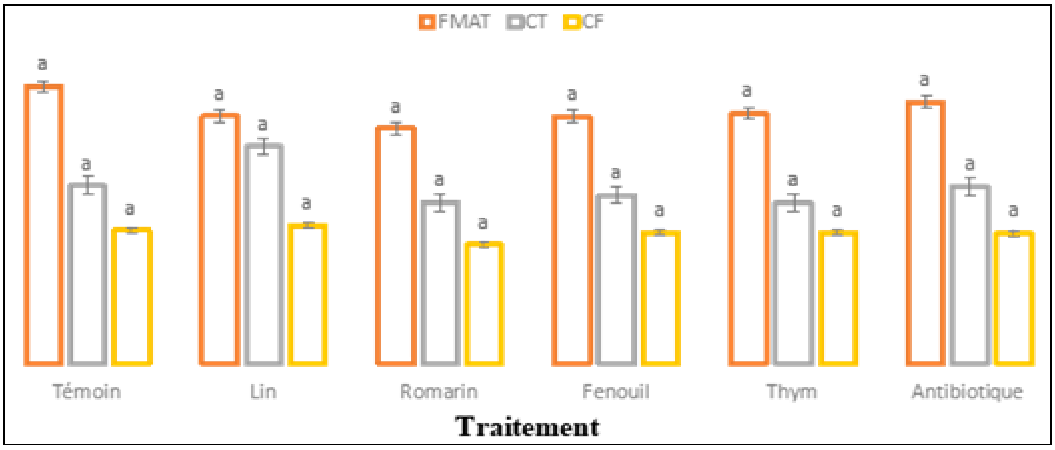

Figure 1: Effect of different treatments on Total Aerobic Mesophilic Flora (FMAT), Total Coliforms (TC) and Faecal Coliforms (FC) in broiler caeca treated with Antibiotic, essential oils of fennel (Fenouil), Linen (Lin), rosemary (Romarin) and thyme (Thym)

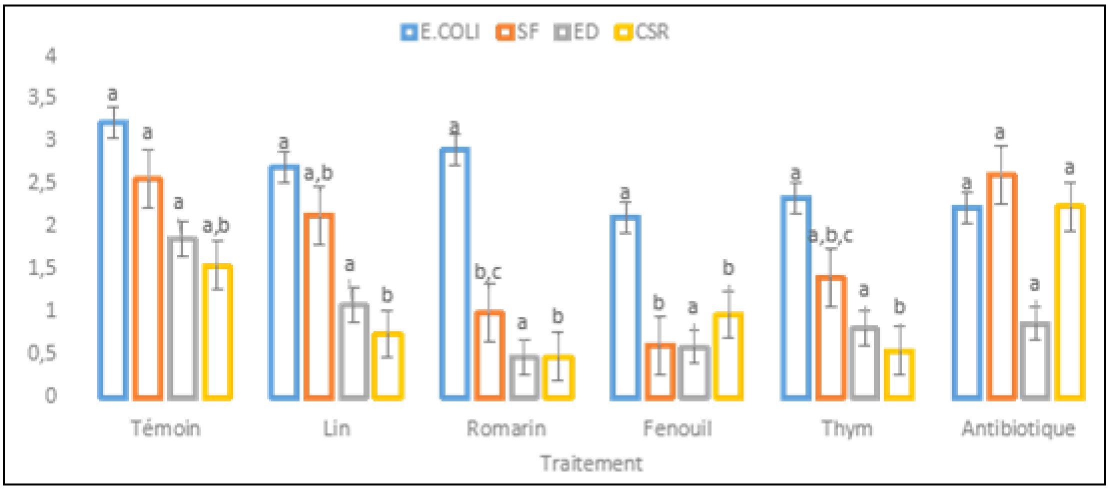

Figure 2: Effect of different treatments on E. coli, S. Fecal (SF), Enterococcus D (ED) and Clostriduimperfirgens (CSR) in broiler caeca treated with essential oils of fennel, linen, rosemary, thyme and antibiotic.

Means with the same letter are not significantly different.

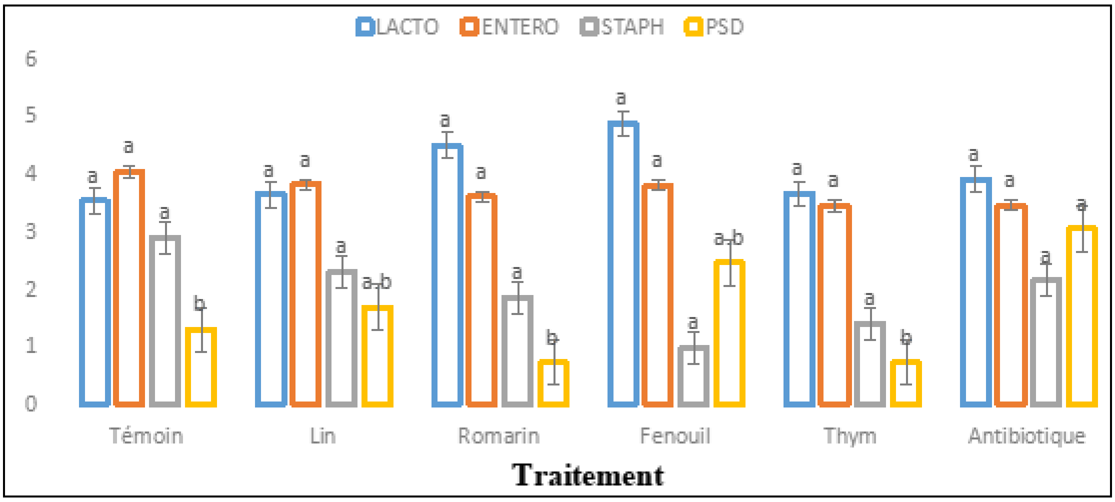

Figure 3: Effect of different treatments on Lactobacillus (LACTO), Enterobacteriaceae (ENTER), Staphillococcus (STAPH) and Pseudominas (PSD) in broiler caeca treated with essential oils of fennel, linen, rosemary, thyme and antibiotic.

Means with the same letter are not significantly different.

3.2. Bacteriological effect on the ileum

With regard to the impact of flax, rosemary, fennel, thyme and antibiotic EOs compared with the control on ileal digestive microbiota, the treatments had a significant impact on SF, PSD and Staph (figs. 25 and 26), which varied according to treatment. The presence of clostridium perfringens was reduced by thyme, fennel and ABT EO compared with the control, while E.Coli concentration was also limited by thyme EO supplementation (fig. 25). The number of bacteria varies (log10UFC/g content).

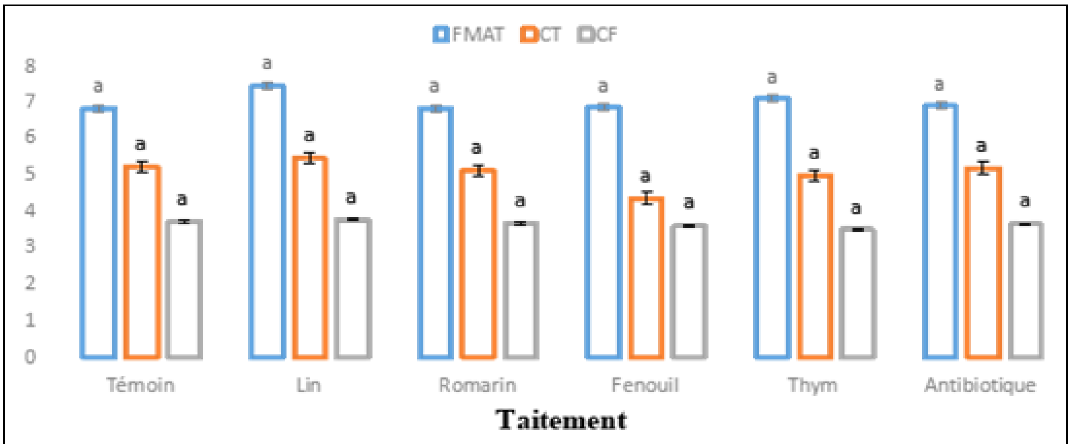

Figure 4: Effect of different treatments on Total Aerobic Mesophilic Flora (FMAT), Total Coliforms (TC) and Faecal Coliforms (FC) concentration in broilerileum treated with Antibiotic, essential oils of fennel (Fenouil), Linen (Lin), rosemary (Romarin) and thyme (Thym)

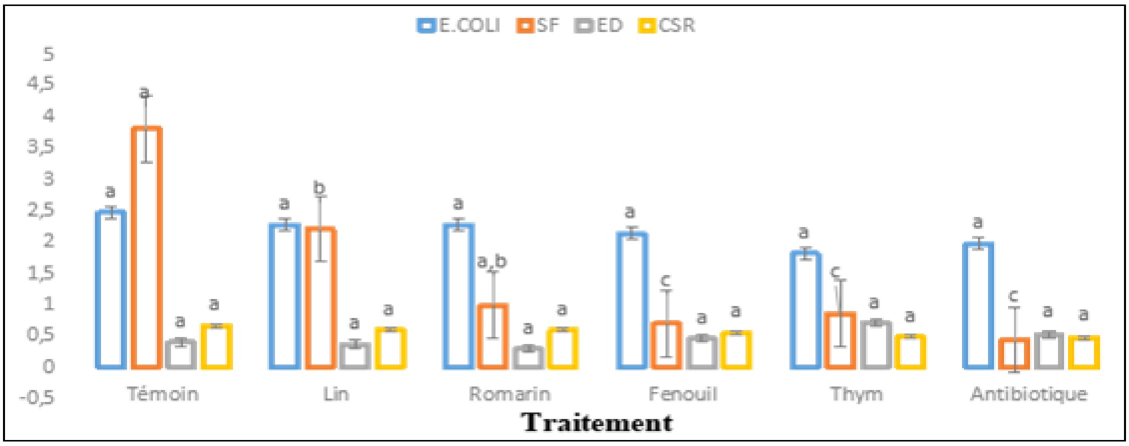

Figure 5: Effect of different treatments on certain pathogenic species in broilers treated with essential oils of fennel, flax, rosemary, thyme and antibiotic.

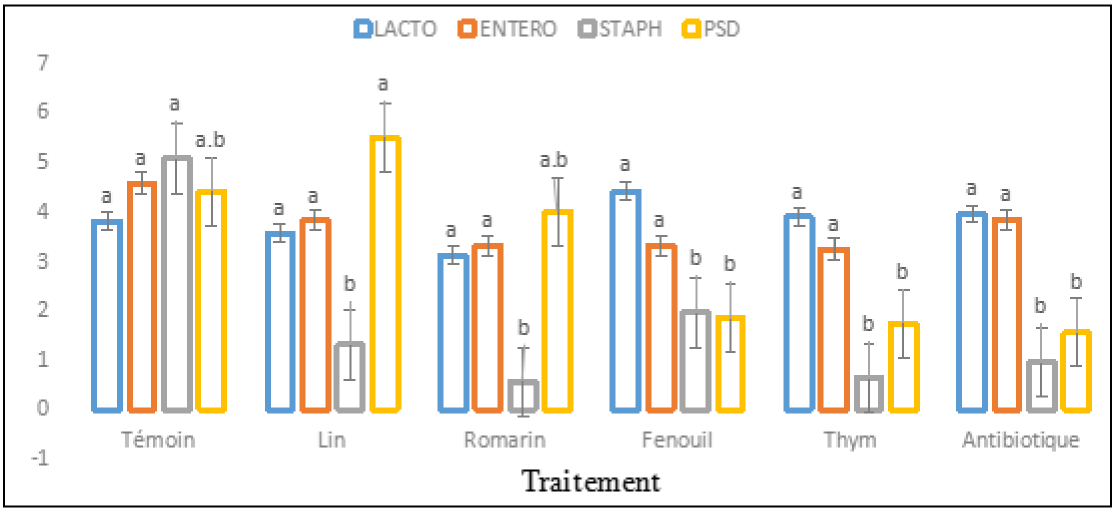

Figure 6: Effect of different treatments on Lactobacillus (LACTO), Enterobacteriaceae (ENTER), Staphillococcus (STAPH) and Pseudominas (PSD) in broilerileum treated with essential oils of fennel, linen, rosemary, thyme and antibiotic.

4. Discussion

The main objective of our study was to determine the effect of essential oils from four aromatic and medicinal plants (fennel, flax, rosemary, thyme) on the digestive microbiota of broiler chickens.

The addition of phytobiotics resulted in a remarkable improvement in the digestive microbiota of poultry, in terms of the impact of the EOs of linen, rosemary, fennel, thyme and the antibiotic compared with the control on the digestive microbiota, the treatments had a significant impact on Total Aerobic Mesophilic Flora (TAMF), Total Coliforms (TC) and Faecal Coliforms (FC) (Figures 1 and 4), which varied according to digestive segment (Ceaca and Iléon). A very limited change was observed for pathogenic species, especially E.coli, C. perfringens and Streptococci in the ileum segments (figure 5), while a significant effect ($p<0.05$) was recorded especially for Clostridium perfringens and Streptococci in the caeca (figure 2). The colonization of Enterobacter (ENTERO), Staphylococcus (STAPH) and Pseudomonas (PSD) species was significantly reduced by these essential oils, especially those from fennel, thyme and rosemary, compared with the control. In vivo studies on poultry suggest that the fenugreek component can modify digestive microflora when added to the diet (Dorman and Deans, 2000).Fennel EO inhibits the growth of Enterococcus and Clostridium perfringens in rat intestine (Liang et al., 2010). Certain components of thyme and rosemary EO, such as thymol, pinene, carvacol and cineol, limit the growth of pathogenic bacteria in the gut (Dorman and Deans, 2000; Bousbia et al., 2009; Imelouane et al., 2009), and are also able to deconjugate bile salts. These constituents appear to act on bacterial membranes by permeabilizing them (Hammer et al., 1999). In addition, the differing effectiveness of these molecules in limiting bacterial growth may be due to their ability to bind hydrogen (Cowan, 1999; Griffin et al., 1999; Ceylan and Fung, 2004; Bakkali et al., 2008). Reducing the microbiotathus increases the availability of certain nutrients to the host. Finally, by limiting the development of the microbiota, they reduce the energy-intensive immune responses associated with it (Humphrey and Klasing, 2004; Windisch et al., 2008).

5. Conclusion

Our results showed that the addition of essential oils to broiler feed had a positive effect on digestive microbiota in the caeca. These phytobiotics revealed a bacteriostatic effect on the bacterial population studied.

These results open up a promising avenue of research into additives with bioactive substances as substitutes for risky conventional products, both for improving the microbial ecosystems of the poultry digestive tract and for enhancing the zootechnical performance of farm animals.

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