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ABSTRACT

In two different goat farms, producing Picodon and Mutschli cheese respectively, two groups of lactating goats were fed alfalfa pellets (ALF) or sainfoin pellets (SNF) for 6 weeks. The cheeses produced at the end of the trial were analysed for sensory properties, composition, fatty acid profile, proteolysis, and rheology. The SNF Picodon had higher “goat” and “farm” aroma and odour, and higher bitter taste, compared with the ALF Picodon. The SNF Picodon showed lower dry matter than the ALF Picodon, leading to differences in texture and mouthfeel. The SNF Mutschli had a different mouthfeel than the ALF Mutschli. In a triangle test, consumers were able to perceive a difference in both Picodon and Mutschli between ALF and SNF groups. In both Picodon and Mutschli, feeding sainfoin increased cheese proteolysis and modified cheese fatty acid profile, resulting in higher polyunsaturated fatty acids proportion and lower *n*-6/*n*-3 ratio.

1. Introduction

Sainfoin (*Onobrychis viciifolia* Scop.) is a legume fodder with high protein content and metabolisable energy (Scharenberg et al., 2007), and it is an interesting choice to reduce chemical inputs and farm environmental footprint and to promote animal health. Indeed, thanks to the ability to fix atmospheric N, sainfoin does not require N fertilisation (Sheehy & McNeill, 1988), thus reducing the demand for chemical inputs in farming. Unlike the widespread alfalfa (*Medicago sativa* L.), sainfoin contains a significant amount of condensed tannin, ranging from 29.7 to 80.1 g kg^{-1} of dry matter (DM) depending on the variety and the growing and harvesting conditions (Azuhnwi et al., 2011; Wang, McAllister, & Acharya, 2015). Thanks to the tannin content, dietary sainfoin naturally decreases the incidence of bloat in ruminants (McMahon et al., 1999), and is a promising strategy to control nematode parasitism (Hoste, Jackson, Athanasiadou, Thamsborg, & Hoskin, 2006). Furthermore, dietary tannins can reduce the ammonia N outflow from rumen, with consequent reduction in urinary N excretion (Herremans, Vanwindekens, Decruyenaere, Beckers, & Froidmont, 2020), and they affect rumen methanogenesis, potentially resulting in a lower emission of CH_4 (Aboagye & Beauchemin, 2019).

Sainfoin could also improve the quality of ruminant products, as tannins are able to modulate the ruminal biohydrogenation (RBH) of fatty acids (FAs) and consequently increase the proportion of potentially healthy FAs in meat and milk (Frutos et al., 2020). The effect of sainfoin on FA profile has been tested in lamb meat (Campidonico et al., 2016; Gruffat, Durand, Rivaroli, Do Prado, & Prache, 2020), ewe milk (Pascual et al., 2019), and cow milk and cheese (Girard et al., 2016; Huyen, Verstegen, Hendriks, & Pellikaan, 2020), resulting in a general increase in polyunsaturated fatty acids (PUFAs), especially *n*-3 series.

However, little is known about the effects of sainfoin on the sensory properties of dairy products. This is an important concern, as sensory properties remain a main driver in consumer choice (Mascarello, Pinto, Parise, Crovato, & Ravarotto, 2015). Pascual et al. (2019) observed no effect of sainfoin hay feeding on the acceptance of ewe curd by untrained panellists, compared with curd produced with milk obtained from ewes fed tall fescue (*Lolium arundinaceum* Darbysh.) hay. Girard et al. (2016) found Gruyère-type cheese obtained from the milk of cows eating sainfoin pellets to be harder and less adhesive, compared with the group eating alfalfa pellets.

To the best of our knowledge, no data are available in the literature about the effects of sainfoin on the overall quality of goat cheese. This is unfortunate, as goat farming can take full advantage of all the benefits of this forage. Our hypothesis was that feeding goats sainfoin would not lend any off-flavour to cheese, while potentially improving its healthiness at the same time. For this purpose, the sensory properties and the FA profile of two different cheese varieties from goats fed sainfoin in replacement of alfalfa were assessed in two different commercial farms, along with other cheese characteristics descriptors. The experiment was designed to directly test the effects of practical sainfoin feeding on farm conditions.

2. Material and methods

2.1. Experimental design, animals, and diets

Two experiments were carried out in the same year during summer grazing in two commercial goat farms, one located in the Drôme department (44.646 °N, 5.040°E, 400 m a.s.l., France), producing Picodon cheese (PCD), and the other located in the Valais canton (46.284 N, 7.882 E, 658 m a.s.l., Switzerland), producing Mutschli cheese (MTS). Animal handling followed the common farming practices.

In both the PCD and MTS experiments, a flock of lactating Alpine goats was divided into two groups, namely alfalfa (ALF) and sainfoin (SNF). In both experiments, the two groups were balanced for milk yield, milk protein content, milk fat content, days in milk, and parity. In each experiment, the ALF goats (n = 14, PCD; 10, MTS) were daily fed with 700 g of alfalfa (*M. sativa*) pellets (8 mm diameter), and the SNF goats (n = 18, PCD; 10, MTS) were daily fed with 700 g of sainfoin (*O. viciifolia* variety Perly) pellets. The sainfoin pellets (8-mm diameter) were produced in a commercial drying and pelleting facility, using the 2nd cut of a pure plot grew in Switzerland (46°85' N, 6°54' E). All animals received a daily supplement of 50 g of whole barley grain or 100 g of corn grit, in the PCD and MTS experiments, respectively. The daily ration of pellets and cereals was split in two equal meals and individually offered twice a day in the milking parlour. The experimental feeding lasted 6 weeks.

Except for milking, all the animals were kept in one flock, in their respective farm, and had access to pasture for 5 h d^{-1}. In the PCD experiment, pasture was composed by 27% tall fescue (*L. arundinaceum*), 27% cock's foot (*Dactylis glomerata* L.), 26% ryegrass and 20% alfalfa. In the MTS experiment, the average biomass coverage of pasture was 40% ryegrass (*Lolium perenne* L.), 20% white clover (*Trifolium repens* L.), 16% various gramineous plants, and 21% other herbs. When stabled, goats had ad libitum access to non-tanniferous hay: pure alfalfa hay (3rd cut, early flower stage) in the PCD experiment, red fescue (*Festuca rubra* L.) and timothy (*Phleum pratense* L.) hay in the MTS experiment.

2.2. Cheesemaking

2.2.1. Picodon

In mid-August, during the last two weeks of experimentation, 6 cheesemaking trials were conducted on different days. On each cheesemaking day, the morning bulk milk of the two experimental groups was collected separately. Within 1 h after the milking, the raw bulk milk of each experimental group was simultaneously processed on farm. The cheesemaking procedure was that of Picodon, a 60-g soft-ripened cheese with bloomy rind made in southern France and granted with a Protected Designation of Origin (French Decree, 2000). Briefly, when the temperature of the milk was equal to 19–21°C, it was seeded with 2% whey from the previous day cheese production and 0.01 % of liquid calf rennet (520 mg L^{-1} of chymosin, Beaugel 500, Etablissements Coquard, Villefranche sur Saône, France). The coagulation lasted 24 h at 22–24 °C, then the curd was carefully poured into individual moulds with a curd ladle without cutting and stirring. The mould draining lasted 24 h, during which time the cheeses were turned over after 6 h and salted with 1 g per cheese of fine solid salt, applied on the upper side. At un-moulding, the other side of each cheese was salted with 1 g of fine solid salt and cheeses were kept in the room for 24 h to promote yeast development on the rind. Then, cheeses were dried on draining grids into a drying room (14 °C, 50% RH) for 5 days. The cheeses were finally ripened in cellars (14 °C, >90% RH) for 14 d and then sampled for analyses.

2.2.2. Mutschli

Between the end of August and the beginning of September, during the last two weeks of experimentation, 4 cheesemaking trials were conducted on different days. On each cheesemaking day, the evening bulk milk of the two experimental groups was collected separately and stored at 4 °C until the next morning. At the morning milking, the bulk milk of the two experimental groups was collected and blended with the respective evening milk. The resulting raw bulk milk of each experimental group was simultaneously processed in Mutschli cheese, a semi-hard pressed traditional Swiss cheese. The raw milk was heated to 62–63 °C for 2–3 min and then cooled to 33–34 °C. Then, it was inoculated with Lyofast MT 092 FET starter culture (1 IMCU per 100 L), and 30 min later with 0.25 mL L^{-1} of BioRen "Piccante" kid rennet (80% chymosin). After 35 min, the curd was cut for 2 min to reach corn kernel-size (6–7 mm) and then blended for 15–20 min, before heating to 43 °C for 50 min. The curd was allowed to settle under the whey, and then gathered into 6–9 moulds (Ø 170 mm) per vat for pressing with a 1 kg weight on each cheese (about 430 Pa) for 5 h. At un-moulding, the cheeses (pH 5.25–5.30) were immersed into a saturated brine for 4 h. Once dried, the cheeses were placed into a ripening cellar (10 °C, 85% RH), where they were turned and washed with salted water daily for 2 weeks and every 2–3 days thereafter. After 8 weeks of ageing, cheeses were weighted and sampled for analyses.

2.3. Physicochemical analyses of feedstuffs, milk, and cheese

2.3.1. Feedstuffs

Crude protein (CP) content was determined in triplicate following the Dumas method (Vario Max Cube, Elementar, Germany). Crude fibre and ash were analysed in triplicate by NIRS (NIR-Flex 500, Büchi, Switzerland). The content of condensed tannins of the experimental pellets was assessed following the method of Grabber, Zeller, and Mueller-Harvey (2013), as reported by Leiber, Arnold, Heckendorn, and Werne (2020). The chemical composition of feedstuffs is shown in Table 1.

2.3.2. Milk

During the cheesemaking period, individual milk production and composition were monitored once a week. Milk samples were analysed for fat, protein, and urea contents (ISO 9622; ISO, 2013) and somatic cells count (SCC; ISO 13366-2; ISO, 2006). The results were then weighted by the individual goat production to obtain the bulk milk composition of the ALF and SNF groups of each experiment.

2.3.3. Cheese

The core pH, DM, and fat content of cheese were measured as reported by Verdier-Metz et al. (2000). Total nitrogen (TN), water-soluble nitrogen (WSN), and phosphotungstic acid-soluble nitrogen (PTSN) were determined following the method described by Ardö (1999). Colour descriptors were analysed on the fresh cut paste surface using a chromameter Minolta CR310 (Minolta France S.A., Carrières-sur-Seine, France) in the L*a*b* space (illuminant D65; 10°

standard observer). Rheology was measured as the strength opposed to uniaxial penetration at constant displacement rate, as described by Lerch et al. (2015). The FA profile of cheese was determined by gas chromatography previous one-step extraction and methylation with methanol/boron trifluoride (95:5, v/v), as reported by Lerch et al. (2015).

2.4. Sensory analyses

Before sensory analyses, the PCD cheeses were vacuum-packaged, stored at –18 °C, and then thawed at 3–5 °C the night before the tasting. The MTS cheeses were stored at 0 °C until the sensory analyses.

2.4.1. Triangle test

A triangle test (ISO 4120; ISO, 2004) was performed on both the PCD and MTS cheeses in a single session, with 38 untrained persons (40% male, 60% female). Participants were goat farmers and their families, all coming from the French region of Drôme. For both the PCD and MTS experiments, three 12 g-samples of cheese, 2 of which randomly belonging to the same dietary treatment (i.e., ALF or SNF), were offered for tasting (serving temperature 20 °C) and participants were asked to identify the odd sample.

2.4.2. Sensory profile

The sensory profile of cheese was evaluated by 10 trained panellists. For both Picodon and Mutschli, 4 training sessions were conducted before the evaluation sessions, to define a sensorial grid according to ISO 13299 (ISO, 2010). The performance of the panel was validated during the training sessions to control repeatability, discrimination capacity, and consensus. For Picodon, 33 sensory attributes were defined: 7 for appearance and texture, 5 for odour, 8 for aroma, 5 for taste, and 8 for mouthfeel. For Mutschli, 32 sensory attributes were defined: 6 for appearance and texture, 4 for odour, 6 for aroma, 6 for taste, and 10 for mouthfeel. A detailed description of all the sensory attributes can be found in Supplementary material Tables S1 and S2.

One cheese for each cheesemaking session and for each treatment (n = 12, PCD; 8, MTS) was used to evaluate the sensory profile. The panel tests were performed in 4 sessions of 1 h each and in 1 session of 1 h 30 min, for PCD and MTS respectively. Cheese samples were served at 21 °C in a monadic sequence, and distributed according to a Williams Latin square design, following ISO 13299 (ISO, 2010) recommendations. Each sensory attribute was evaluated on a 10 cm unstructured line scale, anchored from 0 (= "no perception") to 10 (= "very intense perception"). Data were collected with the Tastel software (version 2011; ABT Informatique, Rouvroy-sur-Marne, France).

2.5. Calculations and statistics

Atherogenicity index (AI) and thrombogenicity index (TI) were calculated according to Ulbricht and Southgate (1991).

Statistical analysis was performed separately for the PCD and MTS experiments. The results of chemical analyses on cheese were statistically elaborated using the mixed ANOVA model of Minitab 19, figuring the fixed effect of treatment (i.e., ALF; SNF) and the random effect of the cheesemaking day. The answers (correct or incorrect) obtained in the triangle test were tested statistically basing on comparisons with values from the binomial law parameter P = 1/3 with n replicates (AFNOR, 1983). The results of sensory profile were analysed using a mixed ANOVA model of SAS 9.4, figuring the fixed effect of treatment (i.e., ALF; SNF) and the random effect of the panellist. Differences were considered significant at $P \leq 0.050$.

3. Results

3.1. Milk composition

In the PCD experiment, the ALF milk and SNF milk had 30.20 and 28.97 g kg^{-1} of fat, 27.69 and 27.65 g kg^{-1} of protein, 32.29 and 33.37 mg dL^{-1} of urea, and 5.58 and 5.27 SCC (log_{10} mL^{-1}), respectively. In the MTS experiment, the ALF and SNF milk had 26.99 and 26.81 g kg^{-1} of fat, 31.10 and 32.22 g kg^{-1} of protein, 66.13 and 61.61 mg dL^{-1} of urea, and 5.41 and 5.75 SCC (log_{10} mL^{-1}), respectively.

3.2. Cheese composition, proteolysis, colour, and rheology

All the results from the analyses of composition, proteolysis, colour, and rheology are shown in Table 2. Sainfoin supplementation increased ($P < 0.050$) the PTSN to WSN ratio in both the PCD and MTS cheeses. In the SNF cheese of the PCD experiment, a lower ($P = 0.013$) DM was observed, along with a lower ($P = 0.039$) Young modulus, compared with the ALF cheese. In the MTS experiment, a lower ($P = 0.045$) redness index (a*) was observed in the SNF cheese compared with the ALF cheese.

3.2.1. Cheese fatty acid profile

The FA profile of the PCD and MTS cheeses is reported in Table 3. In the PCD experiment, dietary sainfoin increased the concentration of 4:0 ($P = 0.020$) and 6:0 ($P = 0.011$) and decreased ($P = 0.002$) the concentration of 12:0 and 14:0 in cheese, without modifying ($P > 0.050$) the total de novo FA concentration. Concerning odd- and branched-chain FAs (OBCFAs), the SNF cheese had lower odd-chain FAs ($P = 0.004$), *iso*-FAs ($P = 0.044$), and *anteiso*-FAs ($P = 0.007$) in the PCD experiment, whereas a reduction in *iso*-15:0 ($P = 0.003$) and *anteiso*-FAs ($P < 0.001$) was observed in the MTS experiment. Feeding sainfoin pellets increased *trans*11-18:1 concentration ($P = 0.009$, PCD; $P = 0.041$, MTS) in both the PCD and MTS experiments, and reduced *cis*9-18:1

concentration ($P = 0.046$) in the MTS experiment. In both experiments, the SNF cheeses showed higher *cis*9*cis*12*cis*15-18:3 concentration ($P = 0.003$, PCD; $P = 0.004$, MTS) and PUFA concentration ($P = 0.004$, PCD; $P = 0.013$, MTS), and lower ($P < 0.001$) *n*-6 to *n*-3 PUFA ratio (*n*-6/*n*-3) compared with the ALF cheeses. In the PCD experiment, sainfoin feeding increased ($P = 0.004$) *cis*9*trans*11-18:2 concentration. A reduction ($P = 0.032$) in TI was observed in the SNF cheese of the PCD experiment compared with the ALF cheese. A more detailed FA profile can be found in Supplementary material Table S3.

3.3. Cheese sensory properties

The participants in triangle test successfully identified the odd cheese ($P < 0.001$), with 78.9% correct answers for the PCD experiment and 60.5% correct answers for the MTS experiment.

The sensory profile of the PCD cheese is reported in Table 4. Panellists found significant differences between the ALF and SNF groups in 15 out of 33 descriptors. The appearance of rind was more homogeneous ($P = 0.050$), less mouldy ($P = 0.048$), and with a thicker underneath layer ($P < 0.001$) in the SNF cheese compared with the ALF cheese. The odour of the SNF cheese was judged more intense ($P = 0.015$), especially considering the farm odour, compared with the ALF cheese. Also, goat and farm aromas were rated more intense ($P = 0.048$, 0.009, respectively) in the SNF cheeses, as well as the bitter taste ($P = 0.009$). Finally, the SNF cheese differed from the ALF cheese in mouthfeel, judged less hard ($P < 0.001$), dry ($P < 0.001$), crumbly ($P = 0.008$) and chalky ($P = 0.038$), and creamier ($P = 0.006$) than the ALF cheese by panellists.

The sensory profile of the MTS cheese is reported in Table 5. Panellists observed significant differences between the ALF and SNF cheeses in 5 out of 32 descriptors. The differences were related to the mouthfeel of cheese: compared with the ALF cheese, the SNF cheese was described as drier ($P = 0.041$), with lower melty ($P = 0.026$), greasy ($P = 0.036$), and sticky ($P = 0.022$) sensations in mouth, and with more ($P = 0.049$) chewing residues.

4. Discussion

4.1. Effect of dietary sainfoin on cheese sensory properties

Feeding sainfoin sensibly modified the sensory properties of Picodon and Mutschli as observed in both sensory profile and triangle test. Interestingly, the differences found by panellists between the ALF and SNF cheeses were sufficient to allow most participants in triangle test to successfully discriminate between the two diets. On the contrary, previous studies on sainfoin feeding reported no, or a limited, effect on the sensory properties of cheese obtained from cow and ewe milk (Girard et al., 2016; Pascual et al., 2019), and Menci et al. (2021) observed no relevant effect of dietary tannin extract on the volatile odour-active compounds of cow cheese. The reason may lie in a species-specific effect due to the different rumen metabolism of goats, ewes, and cows. For example, goats have different ruminal microbiota and RBH pathways compared with cows, and they may react with different mechanisms to dietary challenges such as supplementing fish oil or sunflower oil and starch (Toral et al., 2015, 2016). However, the analyses on milk did not highlight any difference in the main components of goat milk between the ALF and SNF groups.

The effect of dietary sainfoin on cheese sensory properties varied according to the experiment: in the triangle test, the higher percentage of correct responses for the PCD cheeses than for the MTS cheeses revealed a higher impact of sainfoin feeding on Picodon sensory properties. It could also reveal a lower knowledge of Mutschli by the French farmers and their families, who were instead usual consumers of Picodon cheese. Still, the sensory profile confirmed that the impact of sainfoin feeding was limited to mouthfeel in the MTS experiment while it also concerned appearance, odour, aroma, and taste in the PCD experiment. Indeed, the effect of feeding on sensory properties may vary according to the cheese variety: for example, Verdier-Metz et al. (2005) observed that the bigger the cheese size is, the larger the effects are. We observed the opposite here, but the results may be related to different action of sainfoin feeding on cheese according to the cheesemaking process, as discussed later.

4.1.1. Dietary sainfoin effect on cheese gross composition affected cheese texture, mouthfeel, and appearance

Concerning the PCD experiment, the mouthfeel and texture of the SNF cheese were consistent with its rheology and were likely related to the lower DM, compared with the ALF cheese. Indeed, Leclercq-Perlat et al. (2019) previously reported on Picodon cheese that a higher DM content is combined with a higher hardness and a lower creamy and sticky texture. The DM reduction by dietary sainfoin observed in the PCD experiment was not consistent with the MTS experiment, nor with a previous study on cow cheese (Girard et al., 2016). However, this effect was likely due to the sainfoin feeding because the two experimental groups were balanced for milk protein and fat contents. Di Trana et al. (2015) observed an increase in the phenol content of goats' milk after feeding sulla (*Sulla coronaria* Medik.), a tanniferous forage. Phenolic compounds can interact with milk proteins and affect acidification through their antibacterial activity (O'Connell & Fox, 2001). Adding simple phenols (tannic and gallic acids) to acidified milk increased the water binding capacity of gel matrix (Harbourne, Jacquier, & O'Riordan, 2011). Furthermore, a reduced syneresis and a lower DM were observed in cheese fortified with grape phenolic extract (da Silva, Matumoto-Pintro, Bazinet, Couillard, & Britten, 2015). Despite acidification not being monitored during cheesemaking, our observations appear consistent with the literature, suggesting some influence of the phenolic substances potentially present in the SNF milk. These could result from the direct migration of tannins metabolites to milk, or it may be due to the preservation activity of tannins towards plant phenols in the rumen.

The potential effect of dietary sainfoin on cheese acidification might partly explain the different effect observed between the PCD and MTS experiments. Indeed, coagulation relies more on acidification in Picodon than in Mutschli, because of the lower amount of rennet and the lack of a pressing phase in the former. Furthermore, we can hypothesise that the higher temperature reached in the MTS cheesemaking could have selected a microbiota with lower sensitivity to phenolic compounds, compared with the PCD cheese.

In the MTS experiment, gross composition as well as primary proteolysis, known to affect the texture of cheese because of the water-solubility of the primary products of caseins breakdown (Lawrence, Creamer, & Gilles, 1987), did not differ between the SNF and ALF cheeses. The differences in mouthfeel were rather related to cheese FA profile, as the proportion of saturated and unsaturated FAs may affect the texture of cheese (Frétin et al., 2019). In the SNF group, the lower concentration of *cis*9-18:1, the main unsaturated FA, may have increased the melting point and reduced the greasiness of cheese.

The effect observed on the SNF Picodon appearance is controversial, as a lower surface mould development entails a homogenous rind colour (Coppa et al., 2011), but it should result in a thinner under-rind layer. Indeed, Leclercq-Perlat et al. (2019) reported for Picodon cheeses ripened under different temperature and humidity conditions that rind thickness, combined with a higher surface mould (*Geotrichum candidum*) development, is positively correlated with proteolysis and lipolysis, thus affecting the thickness of the under-rind layer. In the present study, the higher PUFA proportion of the SNF cheese may have resulted in a greater oiling off, making rind less suitable for mould development and activity, as hypothesised by Frétin et al. (2019) studying Cantal cheese obtained from the milk of grazing cows. However, this did not prevent the SNF Picodon to undergo a greater proteolysis and exhibit a thicker under-rind layer, probably because the higher moisture of cheese promoted the overall microbial activity. Surprisingly, despite the differences in the appearance observed by panellists between the ALF and SNF Picodon, a visual trait was cited as a motivation for only 2 out of 30 correct responses of triangle test (data not shown).

4.1.2. Dietary sainfoin effect on lipolysis and proteolysis affected cheese odour, aroma, and taste

The more intense goat and farm aroma in the SNF Picodon was likely due to increased lipolysis. Indeed, butanoic, hexanoic, and octanoic acids released by lipolysis contribute to the "goaty" aroma of cheese (Watkins et al., 2021). Although free FAs were not directly analysed in this study, we hypothesised a correlation between such flavour differences and the higher concentration of short-chain FAs found in the SNF Picodon. Conversely, feeding sainfoin did not result in any significant difference in the odour and aroma of Mutschli cheese. This lack of effect was probably due to the differences between the two types of cheese.

The higher secondary proteolysis was probably responsible for the relative bitterness of the SNF Picodon, as the water-insoluble small peptides are known to give a bitter taste (Sousa, Ardö, & McSweeney, 2001). Hypothetically, the bitter taste could also be due to the potential presence of some phenolic compounds transferred from sainfoin to milk, as observed by Di Trana et al. (2015) in goats eating sulla.

Cheese proteolysis has never been assessed in studies feeding ruminants sainfoin or other tanniferous forages. Recently, Menci et al. (2021) found no effect of a dietary tannin extract (from quebracho and chestnut) on the proteolysis indices of grazing cows' cheese. The results of the present study show that dietary tannins from sainfoin may affect proteolysis in goat cheese. In the PCD experiment, the increased PTSN/WSN could reflect the potentially higher microbial activity related to the lower DM, but this does not apply to the MTS experiment. Further research investigating the effect of sainfoin and dietary tannins in general on cheese quality should include the determination of proteolysis to study this aspect.

4.2. Effect of dietary sainfoin on cheese fatty acid profile

The increase in PUFA proportion, as well as the reduction in OBCFA proportion, indicates an impairment of rumen microbiota activity (Frutos et al., 2020), which is consistent with previous studies investigating dietary sainfoin (Girard et al., 2016; Huyen et al., 2020; Pascual et al., 2019).

This reflects the modulation of RBH by tannins contained in the forage: Huyen et al. (2020) found a higher transfer efficiency of *cis*9*cis*12*cis*15-18:3 from feed to milk when replacing 50% of non-tanniferous grass silage in the diet with sainfoin silage. The effect of dietary sainfoin on cheese FA profile observed in the present study is desirable for human diet, as high *cis*9*cis*12*cis*15-18:3, *trans*11-18:1 and *cis*9*trans*11-18:2, and low *n*-6/*n*-3 and TI could reduce cardiovascular disease risk (Gebauer et al., 2011; Harris, 2006; Ulbricht & Southgate, 1991). Considering TI, the potentially beneficial effect of dietary sainfoin was lighter in the MTS than in PCD experiment, probably because of the reduction in *cis*9-18:1 proportion in SNF Mutschli. A reduction in milk *cis*9-18:1 proportion was observed in ewes eating sainfoin (Pascual et al., 2019), sulla (Addis et al., 2005), or supplemented with chestnut tannin extract (Buccioni et al., 2015), whereas an increase in milk *cis*9-18:1 was observed in cows eating buckwheat forage (Kälber, Meier, Kreuzer, & Leiber, 2011) or in ewes supplemented with quebracho tannin extract (Buccioni et al., 2015). As recently reviewed by Frutos et al. (2020), the effect of dietary tannins on milk FA is more consistent for some FA than for others, and the comparison among studies is made difficult by the different tannin sources and doses, animal species and basal diets used.

Finally, the changes in the proportion of some de novo FAs in the SNF Picodon could be due to the increased PUFA flow from the rumen, which had an inhibitory effect on the mammary FAs synthesis (Dorea & Armentano, 2017). However, the proportion of total de novo FAs was not affected by dietary sainfoin in the present study.

The differences between the two experiments were expected because of the known variability among different flocks, different pastures, and the different cheesemaking techniques of Picodon and Mutschli. Moreover, the concentrates offered in the two experiments may have played a role in bringing out such differences, as Pascual et al. (2019) found an interaction between sainfoin hay and different concentrates on the FA profile of ewe milk.

5. Conclusion

The sensory properties of Picodon and Mutschli cheeses were modified by replacing alfalfa with sainfoin in the goat diet. The effect was limited to mouthfeel for Mutschli, whereas it also concerned appearance, odour, aroma, and taste for Picodon. A consumer test should be conducted to determine whether such changes in the intensity of the typical sensory attributes of Picodon and Mutschli reduced the acceptance these cheeses. Picodon cheesemakers have to be aware of the effect of dietary sainfoin on cheese water retention, that could be counterbalanced by increasing the draining phase. Concerning cheese FA profile, although some differences occurred between the PCD and MTS experiments, sainfoin feeding resulted in an overall increase in PUFA proportion and a decrease in *n*-6/*n*-3, which are desirable aspects for human diet. Further studies should investigate the interaction between dietary tannins and cheesemaking technique on cheese gross composition and sensory properties, for example comparing soft and pressed semi-hard cheeses using the same starting milk.

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Table 1

Chemical composition of the experimental pellets and the feedstuffs used in Picodon (PCD) and Mutschli (MTS) experiments.[a]

Item	Sainfoin pellet	Alfalfa pellet	PCD			MTS		
			Barley grain	Hay	Pasture	Corn grit	Hay	Pasture
Dry matter (DM; g $100g^{-1}$)	92.5	91.4	87.2	91.9	19.2	91.0	89.1	18.8
Crude protein (g 100 g^{-1} DM)	18.2	20.1	11.3	15.1	14.6	7.2	11.2	21.9
Crude fibre (g 100 g^{-1} DM)	22.8	27.6	5.4	37.2	34.3	1.9	30.1	21.7
Ash (g $100g^{-1}$ DM	10	14.1	2.5	11	11.2	1.4	8.3	10.2
Condensed tannins (g 100 g^{-1} DM)	4	0.3	na	na	na	na	na	na

[a] Abbreviation: na, not analysed. Data for barley grain and corn grit as provided by the producer.

Table 2

Effect of feeding goats with sainfoin on the composition, proteolysis, colour, and rheology of Picodon and Mutschli cheeses. [a]

Item	Picodon				Mutschli			
	ALF	SNF	SEM	*P*-value	ALF	SNF	SEM	*P*-value
pH	5.218	5.233	0.0355	0.636	6.220	6.085	0.0393	0.082
Dry matter (DM; g $100g^{-1}$)	59.3	55.6	1.32	0.013	55.43	56.16	0.643	0.449
Fat (g $100g^{-1}$ DM)	47.6	49.3	1.02	0.440	41.48	39.50	0.599	0.130
TN (g $100g^{-1}$ DM)	6.81	6.89	0.169	0.770	7.820	8.134	0.0870	0.075
WSN/TN (%)	10.80	11.48	0.595	0.512	18.16	16.98	0.372	0.148
PTSN/TN (%)	5.18	6.53	0.414	0.063	9.64	10.32	0.320	0.188
PTSN/WSN (%)	48.4	56.4	1.75	0.015	53.06	60.74	1.92	0.031
Colour parameters								
L* (lightness)	86.17	85.97	0.517	0.734	82.34	82.95	0.582	0.561
a* (redness)	–0.23	0.18	0.111	0.063	–0.43	–0.82	0.101	0.045
b* (yellowness)	17.59	17.86	0.537	0.810	14.02	14.46	0.244	0.223
Rheology (MPa)								
Strength to 70% deformation	1.05	0.69	0.104	0.095	1.008	1.212	0.0734	0.126
Strength to 90% deformation	1.08	0.72	0.109	0.111	1.170	1.502	0.0966	0.094
Young modulus (undeformability)	1.25	0.82	0.110	0.039	1.31	1.56	0.107	0.268

[a] Abbreviations are: ALF, goats eating 700 g d^{-1} of alfalfa (*Medicago sativa* L.) pellets; SNF, goats eating 700 g d^{-1} of sainfoin (*Onobrychis viciifolia* Scop.) pellets; SEM, standard error of the mean; TN, total nitrogen; WSN, water-soluble nitrogen; PTSN, phosphotungstic acid-soluble nitrogen.

Table 3

Effect of feeding goats with sainfoin on the fatty acid profile (g 100 g^{-1} of FA) of Picodon and Mutschli cheeses. [a]

Item	Picodon				Mutschli			
	ALF	SNF	SEM	*P*-value	ALF	SNF	SEM	*P*-value
4:0	2.032	2.112	0.0167	0.020	1.750	1.829	0.0225	0.059
6:0	2.467	2.558	0.0197	0.011	1.934	1.949	0.0183	0.515
8:0	2.806	2.888	0.0237	0.082	2.108	2.091	0.0245	0.570
10:0	11.06	11.05	0.146	0.980	7.98	7.91	0.121	0.650
12:0	5.67	5.21	0.161	0.002	4.345	4.332	0.0565	0.832
14:0	12.09	11.34	0.199	0.002	11.991	12.248	0.0997	0.175
*cis*9-14:1	0.247	0.214	0.0115	0.012	0.3192	0.3120	0.00386	0.386
15:0	1.718	1.581	0.0343	0.009	1.444	1.419	0.0204	0.056
iso-15:0	0.345	0.300	0.0143	0.019	0.289	0.261	0.0126	0.003
16:0	30.09	30.11	0.419	0.967	29.55	30.26	0.806	0.225
*cis*9-16:1	0.758	0.706	0.0241	0.105	0.791	0.803	0.0183	0.666
17:0	1.236	1.183	0.0121	0.014	1.089	1.056	0.0164	0.078
anteiso-17:0	0.503	0.460	0.0150	0.020	0.437	0.391	0.0162	0.001
18:0	5.01	5.53	0.242	0.117	5.99	6.19	0.200	0.295
*cis*9-18:1	13.53	13.21	0.315	0.422	18.99	17.31	0.555	0.046
*trans*10-18:1	0.1405	0.1399	0.00856	0.970	0.1724	0.1621	0.00368	0.177
*trans*11-18:1	0.794	1.008	0.0597	0.009	1.082	1.224	0.0572	0.041
*cis*9*cis*12-18:2	1.861	1.987	0.0417	0.062	2.235	1.974	0.0895	0.003
*cis*9*trans*11-18:2	0.561	0.663	0.0300	0.004	0.891	0.919	0.0355	0.281
*cis*9*cis*12*cis*15-18:3	1.04	1.72	0.130	0.003	1.13	1.86	0.152	0.004
20:0	0.2548	0.2588	0.00864	0.717	0.2062	0.2070	0.00510	0.922
20:5 *n*-3	0.0730	0.0936	0.00343	<0.001	0.0617	0.0832	0.00484	0.004
22:0	0.1518	0.1505	0.00643	0.848	0.1038	0.1078	0.00514	0.187
22:5 *n*-3	0.2062	0.2453	0.00917	0.002	0.1942	0.2146	0.00859	0.019
22:6 *n*-3	0.0734	0.0806	0.00259	0.113	0.0522	0.0640	0.00342	0.006
24:0	0.0795	0.0860	0.00417	0.126	0.0485	0.0560	0.00300	0.014
Sums and calculations								
de novo FA	66.21	65.27	0.861	0.336	59.7	60.6	1.03	0.314
SFA	76.73	75.94	0.586	0.230	70.31	71.28	0.788	0.236
OCFA	3.835	3.568	0.0557	0.004	3.173	3.125	0.0451	0.514
iso-FA	0.607	0.549	0.0231	0.044	0.597	0.499	0.0333	0.150
anteiso-FA	0.585	0.531	0.0138	0.007	0.523	0.470	0.0170	<0.001
MUFA	16.78	16.65	0.350	0.739	22.68	21.10	0.609	0.056
PUFA	4.11	5.06	0.211	0.004	4.87	5.40	0.202	0.013
n-6/*n*-3 PUFA	1.533	1.065	0.0770	<0.001	1.73	1.00	0.140	<0.001
*cis*9-18:1/16:0	0.446	0.434	0.0827	0.513	0.640	0.564	0.0987	0.090
AI	4.06	3.76	0.151	0.067	2.98	3.19	0.129	0.177
TI	3.33	2.84	0.130	0.003	2.73	2.55	0.110	0.105

[a] Abbreviations are: ALF, goats eating 700 g d^{-1} of alfalfa (*Medicago sativa* L.) pellets; SNF, goats eating 700 g d^{-1} of sainfoin (*Onobrychis viciifolia* Scop.) pellets; SEM, standard error of the mean; FA, fatty acids; SFA, saturated fatty acids; OCFA, odd-chain fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; AI, atherogenicity index (Ulbricht & Southgate, 1991); TI, thrombogenicity index (Ulbricht and Southgate, 1991). *cis*9*trans*11-18:2 coeluted with *trans*7*cis*9-18:2.

Table 4

Effect of feeding goats with sainfoin on sensory profile of Picodon cheese. [a]

Attribute	ALF	SNF	SEM	*P*-value
Appearance and texture				
Rind colour	6.5	6.3	0.09	0.230
Rind colour homogeneity	5.0	5.5	0.16	0.050
Rind roughness	1.3	1.4	0.08	0.526
Rind mouldiness	3.0	2.5	0.16	0.048
Layer under the rind	1.2	2.0	0.09	<0.001
Paste colour	3.0	2.9	0.11	0.573
Paste hardness	7.9	6.9	0.09	<0.001
Odour				
Overall intensity	5.5	5.8	0.09	0.015
Cream	1.7	1.8	0.10	0.668
Mushroom	1.4	1.3	0.09	0.558
Farm	2.2	2.6	0.10	0.010
Yeast	2.1	2.1	0.10	0.940
Aroma				
Overall intensity	6.0	6.2	0.08	0.089
Goat	4.7	5.2	0.08	0.048
Farm	2.1	2.6	0.11	0.009
Green	1.6	1.5	0.09	0.578
Fruit	1.0	0.6	0.07	0.026
Cream	1.9	2.0	0.08	0.253
Mould	1.7	1.6	0.12	0.589
Rancid	2.0	2.1	0.10	0.377
Taste				
Salty	5.8	5.9	0.09	0.298
Sour	3.8	3.9	0.12	0.456
Bitter	2.7	3.1	0.12	0.009
Pungent	2.4	2.7	0.11	0.190
Astringent	2.9	2.9	0.11	0.884
Mouthfeel				
Hard	5.8	4.9	0.13	<0.001
Dry	5.7	4.9	0.13	<0.001
Crumbly	4.6	3.7	0.20	0.008
Creamy	1.1	1.5	0.09	0.006
Chalky	4.4	3.8	0.14	0.038
Sticky	3.1	3.1	0.11	0.982
Doughy	2.8	3.0	0.10	0.436
Chewing residues	1.6	1.4	0.10	0.248

[a] Abbreviations are: ALF, goats eating 700 g d^{-1} of alfalfa (*Medicago sativa* L.) pellets; SNF, goats eating 700 g d^{-1} of sainfoin (*Onobrychis viciifolia* Scop.) pellets; SEM, standard error of the mean.

Table 5

Effect of feeding goats with sainfoin on sensory profile of Mutschli cheese. [a]

Attribute	ALF	SNF	SEM	*P*-value
Paste appearance and texture				
Colour	3.5	4.0	0.18	0.314
Colour homogeneity	7.4	7.4	0.14	0.974
Brightness	1.0	1.0	0.08	1.000
Hardness	6.7	7.0	0.14	0.130
Elasticity	2.4	2.0	0.17	0.258
Greasiness	1.9	1.6	0.10	0.102
Odour				
Overall intensity	6.3	6.3	0.12	0.943
Goat	5.4	5.6	0.18	0.285
Dried forages	1.8	2.0	0.15	0.246
Fermented	3.3	3.3	0.18	0.987
Aroma				
Overall intensity	6.7	6.7	0.11	0.937
Goat	5.5	5.9	0.16	0.074
Dried forages	1.4	1.7	0.14	0.065
Fermented	3.5	3.4	0.17	0.616
Rancid	1.0	1.0	0.12	0.892
Persistency	5.9	6.0	0.14	0.747
Taste				
Salty	6.9	6.7	0.09	0.052
Sour	3.1	3.0	0.15	0.634
Bitter	3.1	3.0	0.15	0.527
Sweet	0.9	0.8	0.10	0.674
Pungent	2.4	2.5	0.16	0.629
Astringent	2.7	2.9	0.17	0.312
Mouthfeel				
Hard	5.3	5.5	0.16	0.314
Dry	4.2	4.6	0.17	0.041
Melty	2.1	1.5	0.17	0.026
Crumbly	2.6	2.6	0.16	0.716
Grainy	2.8	2.9	0.15	0.651
Rubbery	1.5	1.6	0.15	0.813
Greasy	2.4	2.1	0.11	0.036
Sticky	2.0	1.5	0.16	0.022
Doughy	2.3	1.9	0.14	0.188
Chewing residues	2.8	3.2	0.14	0.049

[a] Abbreviations are: ALF, goats eating 700 g d^{-1} of alfalfa (*Medicago sativa* L.) pellets; SNF, goats eating 700 g d^{-1} of sainfoin (*Onobrychis viciifolia* Scop.) pellets; SEM, standard error of the mean.