

Application of Chia Seed Coproduct in Dry-Cured Sausages: Effect upon its Physicochemical Properties [†]

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Abstract: Chia mucilage is a new ingredient with a lot of new applications and so its extraction from seeds generates a great amount of coproducts (CSWM) which composition made it suitable for the meat industry. In this work, the effect of the addition of CSWM and whole chia seeds (WCS) in a dry-cured sausage (DCS) upon its physicochemical properties has been evaluated (4 processing days). Several concentrations of WCS and CSWM (0, 1.5, 3.0, and 4.5%) were added. pH, water activity (*A_w*), residual nitrite level (RNL), the 2-Thiobarbituric acid (TBA) assay and moisture content were determined. WCS and CSWM addition in DCS decreased the pH values in all the samples. For RNL, the values decreased along time without differences between samples. Although all the samples reached at day 4 a weight loss of 30% (industrial criteria for sausages commercialization), any of them reached *A_w* values to be considered as intermediate moisture foods (≤ 0.900). Samples with WCS showed lower TBA values than control. CSWM can be used as an ingredient for DCS showing the same effect upon its physicochemical properties than WCS and so it could be an interesting way to valorize this coproduct.

Keywords: mucilage; chia coproducts; residual nitrite level; intermediate moisture food; dry-cured meat product

1. Introduction

One of the aspects that are characterizing the COVID-19 pandemic and the post-pandemic age, is the great consumers awareness about the consumption of healthier foods [1,2]. This involves practically all types of foods, of which meat and meat products are not the exception. Now, there has been strong pressure for the meat industry to modify its products, making it healthier and in fact, in some sectors of the meat industry itself, they are already working on the development of meat analogues, as the most radical aspect of the sector. There are five diet restrictors, in which, a lot of research is being carried out. International health agencies (FAO, WHO) have mentioned that processed foods should reduce, caloric intake, sugar content, salt concentration, fat content and/or reduce the intake of saturated fats, and diminish, the use of foods additives. In general, the meat industry is not a great sugars “consumer” (especially mono and disaccharides) like another’s, not so of starches, since many meat products include them in their formulation (especially in cooked cured meat products). Salt is essential in the meat products elaboration process and, a minimum concentration is necessary to act as technological agent.

Fats are being replaced by fat substitutes [3]. These ingredients, normally have low calories content, and are reduced in saturated fatty acids. In pandemic era, oleogels are the most studied “fat substitutes”.

Chia (*Salvia hispanica*, L.), has been worldwide accepted by consumers and its presence, as whole seed, whole flour or the inclusion of its oil in the formulation, is very well considered [4]. Therefore, this oilseed, is one of the new ingredients that is being used in new meat products formulation, not only for its excellent nutritional properties, but also for its excellent techno-functional properties, such as water holding capacity (WHC), and oil holding capacity (OHC), among others. The chia processing, has been booming, especially focus for chia oil production. This oilseed is rich in Ω -3 fatty acids. There have also been attempts to obtain industrial chia mucilage, since has very interesting techno-functional properties due to its excellent WHC (among other properties), however, its extraction is technologically complicated and has low yields. From the mucilage extraction process, co-products are generated that could be used to make healthier meat products. The presence of whole seeds in meat products is not new: whole black pepper, pistachios, walnuts or almonds have been incorporated in several meat products.

The aim of this work was to study the effect of the addition of chia seed without mucilage (CSWM) and whole chia seeds (WCS) (at different concentrations) in a dry-cured sausage model system (DCS) upon its physicochemical properties during its elaboration process.

2. Materials and Methods

Dry-cured sausage elaboration process: The composition and elaboration process of DCS was carried out according to the industrial procedure. DCS were elaborated in the IPOA Research Pilot Plant facility. The DCS was prepared according to a traditional formula (only meat percentages add up to 100% and percentages of other ingredients are meat-related): pork lean meat (60%), pork backfat (40%), water (5%), salt (1.8%), ascorbic acid (500 mg/kg), nitrite (100 mg/kg) and spices (0.2% black pepper and 0.01% anise).

The sausages were stuffed into natural lamb casings of 18-22 mm in diameter. Seven batches were prepared: Control (without chia added), WCS (Whole chia seed) added at 1.5%, 3.0 and 4.5% and Chia seed without mucilage (WSWM) added, both types of chia at these concentrations: 1.5%, 3.0 and 4.5%. Chamber drying conditions were as follows: 15 ± 1 °C and $75 \pm 2\%$ relative humidity. After 3 days of dry-curing process, the DCS (WCS and WSWM) were considered ready-to-eat (30% weight losses). The “small” diameter of casings together with their natural origin (lamb small intestine), allows shortened the dry-cured period, and reach the appreciate quality characteristics of this type of DCS.

Moisture content were determined by AOAC (2016) methods [5]. CIELAB (1976) colour parameters: Lightness (L^*), red/green (+/-) co-ordinate (a^*), yellow/blue (+/-) co-ordinate (b^*) were measured using a spectrophotometer Minolta CM Minolta CM-700 (Minolta Camera Co., Osaka, Japan), using D_{65} as illuminant and 10° as standard observer. Psycho-physical magnitudes, Chroma (C^*) and hue (H^*) and colour differences (ΔE^*) were calculated as: $C^* = (a^{*2} + b^{*2})^{1/2}$, $H^* = \tan^{-1}(b^*/a^*)$. Colour differences (ΔE^*) were calculated as: $\Delta E^* = (\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2})^{1/2}$, where $\Delta L^* = (L^*_{\text{sample}} - L^*_{\text{control}})$; $\Delta a^* = (a^*_{\text{sample}} - a^*_{\text{control}})$; $\Delta b^* = (b^*_{\text{sample}} - b^*_{\text{control}})$. Guidelines for meat colour evaluation was followed [6] and Sanchez-Zapata recommendations [7] to determine infinite solid (product thickness) and background was used. The pH of the sausages was measured directly using a Hach puncture electrode probe (5233) connected to a pH-meter (model SensIONTM + pH3, Hach-Lange S.L.U., Vérenaz, Switzerland). The measurement was taken three times, changing the place of electrode insertion. The water activity (A_w) was measured at 25 °C using an electric hygrometer NOVASINA TH200 (Novasina; Axair Ltd., Pfaeffikon, Switzerland). All samples analysis were measured by triplicate, except for colour measurements which 9 measurements, from each sample, were made. Residual nitrite level (mg NaNO_2/kg sample) was determined in agreement with ISO/DIS 2918 standards [8]. Lipid oxidation was assessed in triplicate by the 2-Thiobarbituric acid (TBA) method following the recommendations of Rosmini and co-workers [9]. TBARS values were calculated from a standard curve of malonaldehyde (MA) and expressed as mg MA/kg sample. Samples for analytical measurements (by triplicate) were taken at 0, 1, 2 and 3 days of elaboration process. Three-elaboration process at three different days were made.

The experimental design was according with IPOA-5Stars methodology (healthy, safety, tasty, sustainable and social accepted foods [10].

A Multifactor ANOVA was used to evaluate the influence of chia seed (whole and without mucilage) concentrations (levels: 0, 25, 50 and 75%) and processing time (levels: 0, 1, 2 and 3 days). When differences between levels were found, the Tukey's test was applied.

3. Results

When analysing the colour Lightness (L^*), the effect of WCS and CSWM can be appreciated in Table 1. No significant differences ($p > 0.05$) were found between all the studied concentrations and all curing time. This behaviour would indicate that free water, responsible for a large part of this coordinate in dry-cured meat products, is entrapped in its structure, preventing it, from be available to interact with light (reducing its reflection). For CSWM and control samples, they follow the characteristic evolution of this colour co-ordinate in dry-cured meat products [11].

When analysing the red/green colour co-ordinate (a^*), it can be seen that except on the first day, where the samples with chia, regardless of the type and concentration, the value of this coordinate decreased, however, throughout the elaboration time (Table 1). Between all the samples, we did not show significant differences ($p > 0.05$) between all the samples and processing time. Being able to say that chia did not influence the red component of the colour.

When the values of the yellow/ blue (b^*) co-ordinate were evaluated, it behaves very similar to the L^* co-ordinate (Table 1). In addition, its general behaviour during the dry-curing elaboration process was similar to that described in other raw-cured meat products [10,11]. An analogous behaviour is described for C^* . This indicates that the change in colour saturation will depend on those aspects that mainly affect the b^* colour co-ordinate (C^* is b^* dependent). When hue (H^*) was analysed (Table 1), the values achieved with or without the addition of chia and during the elaboration period of the same, was similar for all samples and these evolutions and, values, were characteristic for these type of meat products [11].

The effect of chia seed type, concentration and processing time upon pH can be observed in Table 1. The results indicated that the chia seed addition, regardless of the concentration and whether or not it had mucilage, decreased this parameter. When analysing the effect of the mucilage present in the seed, it was observed that the decrease in pH was more pronounced in the samples with mucilage. However, in these samples, the evolution of the pH throughout the elaboration process was not significant ($p > 0.05$) in the samples added with 3 and 4.5% WCS. The effect of the mucilage seems to have an influence on maintaining the pH values (like a buffer) during the manufacturing process (acting upon the effect of lactic acid released by microbiota metabolism over meat matrix, looks like the "acids" are included upon mucilage structure). This effect has been seen with the adsorption of coloured substances in the structure of the mucilage (unpublished data). This aspect may have to be considered in the DCS elaboration process, since pH plays an important role upon sensorial, technological and industrial properties. When analysing the effect that the incorporation of chia seeds without mucilage, it can be seen that depending on the concentration, the decrease in pH values were greater at higher added concentration. During the elaboration process, the pH values decreased in all the samples, no significant differences ($p > 0.05$) were found between the three CSWM samples at the end of the DCS elaboration process, they were even lower than the control. That is, in a way that would be good, enhancing the action of the microbiota or releasing an "acidic substances" from the chia seed. The values reached at the end of the drying process are similar to others reported for similar products [12,13]. To reduce the pH levels of a meat product similar to this, in a "natural way", the use of chia seed without mucilage could be a good alternative to glucono-delta-lactone (GDL, E-575).

When the A_w was analysed (Table 1), it can be seen that regardless of the type and concentration of chia added, this parameter decreased during the dry-curing process in all samples (including the control). When analysing, independently, each one of the “chia seeds”, it can be seen how the water activity values decreased more, in the WCS samples, than in the samples with seed without mucilage. This behaviour would indicate that the mucilage is reducing the availability of water, caused by the hygroscopicity of the chia mucilage. The A_w values for all these samples, including the control, decrease throughout the processing time. Characteristic appearance of this type of sausage and drying process. When analysing CSWM samples, the A_w values were lower, indicating that the lack of mucilage did affect the evolution of the product and, at the end of elaboration process, similar values to the control were founded. The mucilage present in the seeds can be an auxiliary to reduce water activity and thus to reduce the processing time in DCS with higher diameters (55 mm, 65 mm) [13].

Table 1 shows the results of residual nitrite level (RNL). The results showed that the addition of chia seeds, regardless of the form and concentration added, decreased the values of this parameter. The evolution of the decrease in nitrites throughout the production process was similar to other dry-cured meat products, but not the final concentrations reached. In the samples of WCS they present higher values of RNL compared to CSWM. These lower values could be due to the fact that the compounds rich in polyphenols of the external chia seed peel, which in the samples with mucilage, would be protecting from these reactive substances. When analysing the TBA values (Table 1), it can be seen that on average, the incorporation of chia seeds, regardless of the type and concentration added, the TBA values were higher than the control. However, it is appreciated that the WCS, presented lower TBA values than the other samples that is, it would be acting, in a certain way as an antioxidant in the meat matrix. CSWM samples, showed higher TBA values, perhaps due to the treatment they have undergone after extraction of the mucilage and subsequent drying at 65 °C for 1 day.

In Table 1, moisture content of all experiment can be observed. As can be seen, on the last day of the elaboration process, the differences in moisture content can be really appreciated. In third day of processing, WCS samples showed the lowest moisture values. This behaviour indicated that WCS could help the dry-curing process, and could be an auxiliary ingredient, to reduce costs in the dry-curing process.

Table 1. Mean and standard deviation of CIELAB colour parameters (L*: lightness; a*: red/green co-ordinate (+/-); b*: yellow/blue co-ordinate (+/-); C*: Chroma; H*: hue), pH, water activity (Aw), residual nitrite level (RNL), 2-Thiobarbituric acid assay (TBA) and moisture (Moist.) of dry-cured sausages added with different concentrations (0, 1.5, 3.0 and 4.5%) of chia whole seed (CWS) and chia seed without mucilage (CSWM) during a traditional dry-curing elaboration process (0, 1, 2, 3 days).

| Days | CSWM | | | | WCS | | | |
|------|---------|------------------|------------------|-----------------|------------------|-----------------|------------------|------------------|
| | Control | 1.5% | 3% | 4.5% | 1.5 % | 3 % | 4.5% | |
| L* | 0 | 44.07 ± 0.23Aa | 44.49±1.27 Aa | 43.42±0.27Aa | 41.29±0.76 Aa | 50.24± 0.13Aa | 48.04±1.06Aa | 46.28 ± 1.41Aa |
| | 1 | 54.43 ± 3.82Aa | 51.33±2.79 Aa | 48.04±3.45Aa | 46.62±2.26Aa | 47.46± 4.00 Aa | 47.20±1.75Aa | 46.92 ± 3.00Aa |
| | 2 | 48.93 ± 3.41Aa | 47.46±4.00Aa | 46.22±2.20Aa | 44.89±4.55Aa | 46.32±5.67Aa | 45.25±2.85Aa | 45.82 ± 4.17Aa |
| | 3 | 45.39 ± 2.61Aa | 48.67±2.87Aa | 46.61±6.97Aa | 43.88±2.96Aa | 46.18±5.56Aa | 45.48±2.53Aa | 45.84 ± 5.17Aa |
| a* | 0 | 3.50 ± 0.07Aa | 1.61 ± 0.22Aa | 2.08 ± 0.37 Aa | 2.21 ± 0.12 Aa | 3.64 ± 0.07 Aa | 2.35 ± 0.46 Aa | 1.75 ± 0.22 Aa |
| | 1 | 4.62 ± 1.75Aa | 4.49 ± 1.05 Aa | 4.91 ± 1.55 Aa | 4.50 ± 0.95 Aa | 4.46 ± 1.49 Aa | 4.75 ± 1.04 Aa | 3.59 ± 0.58 Aa |
| | 2 | 4.27 ± 1.08Aa | 4.46 ± 1.49Aa | 3.36 ± 1.13 Aa | 3.10 ± 1.55 Aa | 3.99 ± 1.61 Aa | 3.77 ± 0.78 Aa | 3.05 ± 1.28 Aa |
| | 3 | 5.50 ± 0.72Aa | 3.27 ± 1.35b | 3.02 ± 1.66b | 3.32 ± 0.87b | 3.96 ± 1.45b | 3.09 ± 1.43b | 3.26 ± 1.37b |
| b* | 0 | 6.50 ± 0.23Cab | 6.11 ± 0.43Cb | 5.65 ± 0.82Cbc | 5.66 ± 0.14Dc | 6.59 ± 0.08Ba | 6.88 ± 0.20Ba | 5.59 ± 0.48Bc |
| | 1 | 10.04 ± 1.30Aa | 9.22 ± 0.52Aab | 9.08 ± 1.31Aab | 8.05 ± 1.52Abc | 7.02±2.64ABbcd | 7.76 ± 1.02Acd | 7.41 ± 1.52Acd |
| | 2 | 8.02 ± 1.84ABa | 7.02±2.64ABab | 6.16 ± 1.14Bb | 7.23±1.70ABab | 6.71±1.67ABabc | 6.71±1.72ABabc | 6.0 ± 1.30ABabc |
| | 3 | 8.05 ± 1.36ABa | 7.84±1.18ABab | 5.79±1.75ABbc | 6.51±1.52ABCbc | 5.44±1.81ABCbc | 6.56±2.40ABab | 4.41 ± 0.92Ccd |
| C* | 0 | 7.65 ± 0.23Cb | 6.32 ± 0.45Cc | 5.09 ± 0.90Cd | 6.07 ± 0.10BCc | 10.25 ± 0.08Aa | 7.28 ± 0.30Bbc | 5.86 ± 0.49Bb |
| | 1 | 12.41 ± 1.60Aa | 10.28±0.74Aab | 10.38±1.64Aab | 9.23 ± 1.74Ab | 8.43±2.64ABbc | 9.99 ± 1.27Ab | 9.17 ± 1.41Ab |
| | 2 | 9.18 ± 1.63Ba | 8.43±2.64ABab | 7.06±1.38ABab | 7.9 ± 2.12ABbc | 8.02 ± 1.24Bbc | 7.75±1.54ABbc | 6.89 ± 1.48ABbc |
| | 3 | 9.81 ± 1.08Ba | 8.57 ± 1.27Bab | 6.67±1.95BCbc | 7.37 ± 1.42Bbc | 6.82±1.99BCbc | 7.36 ± 2.64 | 5.65 ± 0.83Bc |
| H* | 0 | 62.76 ± 0.63Be | 75.29 ± 1.55Aa | 65.89 ± 1.04Ad | 68.57 ± 1.53Ac | 69.21±0.37Abc | 71.22±3.15Aab | 72.66 ± 1.83Ab |
| | 1 | 68.51 ± 6.98Aa | 64.24±5.05ABCb | 61.9 ± 6.59Abc | 60.75 ± 2.67Bc | 56.94±10.38Bbc | 61.74±4.20Bbc | 66.40 ± 5.05Bbc |
| | 2 | 61.33 ± 8.52ABab | 67.94±10.38ABab | 61.78 ± 6.93Ab | 68.27±7.19ABab | 58.98±14.51Bab | 59.65±8.54Bab | 63.82 ± 9.38Bab |
| | 3 | 55.26 ± 6.59BCa | 56.37 ± 8.25Ca | 58.70± 13.59Aab | 56.25±7.88BCab | 53.48±10.94BCab | 58.16±10.68Bab | 51.25±14.76BCab |
| pH | 0 | 5.78 ± 0.02Aa | 5.74 ± 0.02Ab | 5.68 ± 0.01Ac | 5.63 ± 0.01Ad | 5.63 ± 0.02Ad | 5.64 ± 0.03Ad | 5.63 ± 0.02Ad |
| | 1 | 5.76 ± 0.02Aa | 5.73 ± 0.01Aab | 5.64 ± 0.02Ac | 5.64 ± 0.02Ac | 5.61 ± 0.03Abc | 5.62 ± 0.02Ac | 5.63 ± 0.02Ac |
| | 2 | 5.61 ± 0.03Ba | 5.50 ± 0.01Bb | 5.52 ± 0.02Bb | 5.55 ± 0.03Bbc | 5.59 ± 0.02Acd | 5.61 ± 0.01Ad | 5.61 ± 0.01Ad |
| | 3 | 5.58 ± 0.03Bb | 5.42 ± 0.03Cc | 5.42 ± 0.02Cc | 5.44 ± 0.02Cc | 5.58 ± 0.03Ab | 5.99 ± 0.04Aa | 5.60 ± 0.04Ab |
| Aw | 0 | 0.989 ± 0.004Aa | 0.984 ± 0.010Aab | 0.985 ± 0.002Aa | 0.979 ± 0.001Ab | 0.969 ± 0.001Ac | 0.968 ± 0.023Ac | 0.965 ± 0.003Acd |
| | 1 | 0.974 ± 0.003Ba | 0.948 ± 0.002Bb | 0.942 ± 0.002Bc | 0.940 ± 0.001Bc | 0.941 ± 0.001Bc | 0.939 ± 0.002Bcd | 0.935 ± 0.001Be |
| | 2 | 0.939 ± 0.004Ca | 0.942 ± 0.001Ca | 0.939 ± 0.001Ca | 0.937 ± 0.002BCa | 0.923 ± 0.002Cc | 0.932 ± 0.002Cb | 0.932 ± 0.001Cb |

| | | | | | | | | |
|------------------------------------|----------|-----------------|-----------------|-----------------|------------------|-----------------|-----------------|-----------------|
| | 3 | 0.931 ± 0.002Db | 0.938 ± 0.003Da | 0.933 ± 0.003Db | 0.929 ± 0.003Dbc | 0.92 ± 0.003CDc | 0.927 ± 0.002Dd | 0.926 ± 0.002Dd |
| RNL mg/kg | 0 | 99.05 ± 1.53Aa | 79.17 ± 0.32Ac | 82.08 ± 1.13Ab | 65.93 ± 1.54Ad | 79.04 ± 0.82Ac | 80.19 ± 0.4Ac | 79.76 ± 0.97Ac |
| | 1 | 86.47 ± 1.08Ba | 62.32 ± 0.70Bc | 63.45 ± 1.76Bc | 65.23 ± 0.36Ab | 66.15 ± 0.54Bb | 62.88 ± 0.08Bc | 62.29 ± 0.82Bc |
| | 2 | 71.49 ± 0.78Ca | 57.39 ± 1.65Cd | 41.81 ± 0.21Ce | 50.03 ± 0.07Bd | 53.66 ± 0.67Cc | 53.45 ± 0.62Cc | 58.47 ± 0.88Cb |
| | 3 | 31.59 ± 0.15Da | 7.14 ± 0.42Dd | 3.00 ± 0.05Df | 5.25 ± 0.54Ce | 14.85 ± 0.36Db | 13.17 ± 0.47Dc | 13.84 ± 0.01Dbc |
| TBA mgMA/kg | 0 | 0.16 ± 0.08Aa | 0.19 ± 0.02Aab | 0.13 ± 0.10Aa | 0.17 ± 0.02Aa | 0.20 ± 0.09Ab | 0.20 ± 0.09Ab | 0.19 ± 0.02Ab |
| | 1 | 0.27 ± 0.06Ba | 0.29 ± 0.12Bb | 0.35 ± 0.03Babc | 0.39 ± 0.06Bc | 0.25 ± 0.06Aab | 0.36 ± 0.13Bc | 0.37 ± 0.11Bc |
| | 2 | 0.54 ± 0.16Cab | 0.58 ± 0.03Ca | 0.67 ± 0.14Cb | 0.61 ± 0.05Cb | 0.55 ± 0.03Ba | 0.51 ± 0.26BCab | 0.48 ± 0.10BCa |
| | 3 | 0.79 ± 0.04Db | 0.90 ± 0.07Da | 0.84 ± 0.01Da | 0.78 ± 0.16Dab | 0.63 ± 0.06Cc | 0.66 ± 0.11Cc | 0.69 ± 0.08Dc |
| Moist. gH₂O/100g | 0 | 64.28 ± 0.65Aa | 63.23 ± 1.29Aa | 64.02 ± 0.56Aa | 66.22 ± 3.92Aa | 64.67 ± 1.11Aa | 64.40 ± 0.62Aa | 64.54 ± 0.33Aa |
| | 1 | 60.09 ± 1.62Ba | 59.28 ± 0.43Ba | 59.89 ± 1.53Ba | 62.72 ± 0.48ABb | 61.14 ± 1.40Ba | 60.45 ± 1.28Ba | 60.07 ± 0.38Ba |
| | 2 | 54.77 ± 0.69Ca | 54.75 ± 1.16Ca | 56.92 ± 1.59Ca | 52.75 ± 1.03Cb | 56.57 ± 0.39Ca | 57.84 ± 5.89BCa | 53.42 ± 0.83Cb |
| | 3 | 42.48 ± 2.18Da | 41.12 ± 0.67Da | 35.15 ± 1.65Db | 40.14 ± 1.71Da | 34.29 ± 2.95Db | 35.12 ± 1.59Db | 33.13 ± 1.45Db |

^{A-D} Similar values in the same column indicates not significant differences ($p > 0.05$); ^{a-g} Similar values in the same row indicates not significant differences ($p > 0.05$); MA: malonaldehyde.

4. Conclusions

This work opens a very interesting door to further studies in which chia seeds (as whole chia seed and as its coproduct obtained after mucilage extraction) can be applied in conventional formulations and broadening the knowledge of the interesting technological advantages, that chia could bring to this type of meat products to make them healthier and more attractive to the consumer. The results suggest that whole chia seed (WCS) at any of the concentrations (1.5, 3.0 and 4.5%) under study, is a very good option for dry-cured sausages elaboration process.

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