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CHEMICAL and MICROBIAL DIFFERENCES in DRIED APRICOTS CONTAINING SULFUR DIOXIDE at DIFFERENT LEVELS

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Abstract

This study was conducted to evaluate the effects of different sulfur dioxide (SO_2) concentrations (0, 188, 452, 791, 1034, 1236, 2899 and 3864 mg SO_2/kg) on the physical, chemical and microbiological qualities of dried apricots (DAs). Non-sulfited DAs were evaluated as a control group. Strong correlations were determined between SO_2 concentration with pH (r = – 0.913), titratable acidity values (r = 0.983), browning values (r = – 0.981) and β-carotene contents (r = 0.822) of DAs (P<0.05). The number of total psycrophilic aerobic bacteria (TPAB), lactic acid bacteria, yeast and mold, xerophilic mould, *Staphylococcus* spp., and total *Enterobacteriaceae* were below the detection limit (<0.6 log cfu/g) in samples containing SO_2 even at the lowest level (188 mg SO_2/kg). Although 188 mg SO_2 per kg of DAs is sufficient SO_2 concentration with respect to browning color and microbial load right after drying, DAs should contain minimum 791 mg SO_2/kg to protect the initial β-carotene content.

Keywords: Dried apricots, SO_2 level, browning, β-carotene, microbial load

FARKLI DÜZEYDE SO_2 İÇEREN KURU KAYISILARDAKİ KİMYASAL ve MİKROBİYEL FARKLILIKLAR

Özet

Bu çalışma; kuru kayısıların, fiziksel, kimyasal ve mikrobiyolojik özellikleri üzerine farklı kükürt dioksit (SO_2) konsantrasyonlarının (0, 188, 452, 791, 1034, 1236, 2899 and 3864 mg SO_2/kg) etkisini değerlendirmek amacıyla yürütülmüştür. Kükürt içermeyen kuru kayısı örnekleri kontrol grubu olarak değerlendirilmiştir. Kuru kayısıların içerdiği SO_2 konsantrasyonu ile pH (r = – 0.913), titrasyon asitliği (r = 0.983), esmerleşme değerleri (r = – 0.981) ve β-karoten miktarları (r = 0.822) arasında güçlü korelasyonlar tespit edilmiştir (P<0.05). En düşük düzeyde SO_2 içeren (188 mg SO_2/kg) örnekte dahi toplam psikrofilik bakteri, laktik asit bakterisi, *Staphylococcus* spp., toplam *Enterobacteriaceae*, kserofilik küf ile toplam küf ve maya sayıları tespit limitlerinin (<0.6 log kob/g) altındadır. Kurutma sonrasında 188 mg/kg düzeyindeki SO_2 konsantrasyonu, esmer renk ve mikrobiyel yük açısından yeterli olmasına rağmen, başlangıçtaki β-karoten miktarını korumak için gerekli olan en düşük SO_2 konsantrasyonu 791 mg/kg'dır.

Anahtar kelimeler: Kuru kayısı, SO2 düzeyi, esmerleşme, β-karoten, mikrobiyel yük

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INTRODUCTION

Turkey is the leader in the world for fresh and DA production (1). Almost all of the DA production is obtained in Malatya, a city located eastern part of Turkey. Turkey produces approximately 20% of the world fresh apricot (716000 metric tons, Mt) and 84% of the DA (98000 Mt) (1). DA is the second major dried fruit export product of Turkey, after Sultana raisins. According to the statistical data in 2010, the primary importer countries of Turkish DAs are USA, Russia, England, Germany, France and Australia with a share of about 60% (2).

Commercially produced DAs are usually sulfured for both inhibiting the enzymatic browning during drying and non-enzymatic browning reactions during both drying and subsequent storage. Other than its well-known effects on browning reactions, SO_2 in DAs also acts as: 1) an antioxidant preventing oxidative degradations, 2) an inhibitor of some enzymes such as oxidases, proteases and peroxidases, 3) an antimicrobial agent preventing the growth of especially yeasts and molds (3) and 4) plasmolyzing cells (4), which facilitates drying (5).

The maximum limit of 2000 mg SO_2 per kg of DAs is accepted by most countries, including Turkey (6). The current SO_2 concentration in dried fruits does not possess potential risk to the majority of the population (7, 8). However, the sensitivity of some people to SO_2 has drawn public attention over the safety of sulfites in foods. A study in 1985 by the Federation of American Societies for Experimental Biology indicated that ingesting sulfites might induce to an acute and sometimes life-threatening attack of asthma in some patients (9). According to FDA, one in 100 people is sulfite-sensitive to some degree, but for the 10% of the population who are asthmatic, up to 5 percent are at risk of having an adverse reaction to sulfites (9). Due to the concern about the safety of sulfites, FDA revoked GRAS (generally recognized as safe) status of sulfites for use in fresh fruits and vegetables in 1986 (10).

Taking into consideration the current and probably future restrictions to the uses of sulfites in foods, researchers have studied to develop the sulfite substitutes. Unfortunately, to date, sulfite substitute as functional as sulfite itself could not have been developed. Thus, finding the minimum SO_2 concentration to protect the chemical as well as microbial qualities of DAs is very important. In the present study, the effects of different SO_2 concentrations on the physical, chemical and microbiological qualities of DAs were evaluated. Moreover, it is also our aim to find out the minimum SO_2 concentration which was sufficient for protecting the color and the preventing the microbial growth of DAs.

MATERIALS and METHODS

Materials

Apricots (*Prunus armenica* L., var. Hacıhaliloğlu), provided by the Institute of Fruit Research Center, Malatya, were harvested in July 2008. Hacıhaliloğlu cultivar is the most commonly grown cultivar in Malatya and is very suitable for drying due to its high soluble solid content (°Brix) which is between 24 and 28. In Malatya, 70% of the apricots destined for drying belong to this cultivar.

Methods

Sulfuring and Sun-drying

After visually sorting for sound fruits, fresh fruits (120 kg) were placed in wooden crates (90 x 180 cm) and the crates were stacked on top of each other in the sulfur house. Then, the apricots were sulfured by burning of elemental sulfur. Various sulfur doses and exposure times were applied (Table 1). After exposing the apricots to SO_2 gas, the sulfited apricots in crates were removed from the sulfur house and placed under direct sunlight. On the 3th day of drying, the pits were removed by squeezing the fruit by hand. At the end of 6 days of drying, the drying process was terminated.

As seen in Table 1, the amount of elemental sulfur burned for the first 5 sulfuring trials was the same (500 g). These 5 trials, in which samples contained SO_2 from 188 to 1236 mg per kg, were carried out by the authors. For the last two trials, in which samples contained 2899 and 3864 mg SO_2/kg, the sulfuring was carried out commercially; therefore, the amount of sulfur burned was different than our sulfuring trials. Although they used almost half of the sulfur we used in our sulfuring trials, they exposed to apricots to SO_2 gas in sulfur house at much longer times; i.e., 10 h for the DAs contained 2899 mg SO_2/kg and 12 h for the samples contained 3864 mg SO_2/kg. The

Table 1 Sulfuring parameters of apricots and the final SO_2 concentration in sulfited-dried apricot samples

The number of sample	Sulfur powder content (g)	Sulfuring time (h)	SO_2 concentration of dried apricots (mg/kg)
0	0	0.00	0
1	500	0.50	188
2	500	1.00	452
3	500	1.25	791
4	500	1.50	1034
5	500	2.00	1236
6	240	10.00	2899
7	240	12.00	3864

last two samples which contained the highest amount of SO_2 were included at the later stage of the study. Therefore, we had to take these samples from one of the major producers of DAs in Malatya.

Sampling

The sulfited and sun-DAs were then brought to Ankara University Food Engineering Department where all physical, chemical and microbiological analyses were carried out. The bulk samples at each sulfur level were mixed thoroughly and left in enclosed containers at 20 °C for 2 weeks to equilibrate moisture content. Then, the damaged apricots were selected and removed.

Moisture Analysis

The moisture content of sulfited-DAs (SDAs) was determined in quadruplicate using a vacuum oven (Heraeus VT 6025, Hanau, Germany) at 70 ± 0.1 °C for 14 h by the method (934.06) outlined by AOAC (11).

Water Activity Measurements

The water activity (a_w) of SDAs was measured at 25 °C with a hygrometer (AquaLab 3, Decagon Devices, Pullman, WA) with an accuracy ±0.003.

Sulfur Dioxide Analysis

Sulfur dioxide (SO_2) was determined in duplicate by the modified Monier Williams distillation method (12) with the minor revision in collecting the SO_2 gas described by Franzke et al. (13). The SO_2 gas formed after heating of sulfited sample in a distillation flask with 15% (v/v) HCl was collected into two receiver tubes containing 3% (v/v) H_2O_2. By this way, if the SO_2 gas escaped from the first receiver tube, then it was collected in the second one. The contents of two receiver tubes were combined and then titrated with standardized 0.1 N NaOH. The SO_2 contents of SDAs were expressed as mg/kg.

Browning Measurements

Browning was measured in duplicate according to the method developed for dried carrots by Baloch et al. (14) with minor revision in sample preparation described by Özkan and Cemeroğlu (15). Extraction of the water soluble brown pigment was carried out with 20 ml/L acetic acid containing 10 ml/L formaldehyde. Interfering carotenoid pigments were removed with lead acetate and ethyl alcohol. Formaldehyde was used to remove the interfering SO_2. Absorbances of supernatants were recorded at 420 and 600 nm, using an UV-VIS double-beam spectrophotometer (ThermoSpectronic Helios-α, Cambridge, England). The browning was calculated by subtracting absorbances at 600 nm (for turbidity) from those of 420 nm. The results were expressed as *"absorbance at 420 nm/g sample dried weight."*

pH and Titratable Acidity

Ten grams (±0.01 g) (Sartorius BP 3100S, Goettingen, Germany) of homogenized sample were rehydrated with 90 g of distilled water at 4 °C overnight. The mixture was then homogenized for 5 min in a high-speed stainless steel blender (Waring 8012, Torrington, CT) and the resulting homogenate was filtered through cheese cloth. The filtrate was used for both pH and titratable acidity analyses. pH was measured with a pH meter (WTW Inolab Level 1, Weilheim, Germany). Titratable acidity was determined according to the method (942.15) outlined by AOAC (11) and expressed as *"g anhydrous citric acid/100 g sample."*

β-carotene Analysis

Extraction

β-carotene was extracted following the method described by Sadler et al. (16) with minor revisions in sample preparation before extraction. A 15 ± 0.01 g of minced sample was rehydrated in 45 mL

distilled water at 4 °C overnight. This mixture was homogenized first in the Waring blender for 2 min and then in the homogenizer at 13500 rpm for an additional 2 min to obtain a thoroughly homogenized sample. A 5 ± 0.0001 g of homogenized sample was precisely weighed directly into a polypropylene centrifuge tube using an electronic balance (Mettler Toledo XS 205, Greifensee, Switzerland). Calcium carbonate (0.5 g) was added as a neutralizing agent. An extraction solvent of 20 mL, consisting of hexane:acetone:ethanol (50:25:25, v/v/v), was added to the centrifuge tube which was agitated on a orbital shaker (Heidolph Unimax 2010, Schwabach, Germany) at 220 rpm until the residue became completely colourless (app. 15 min). A 5 mL of distilled water was added to hasten the phase separation, followed by centrifugation at 9400xg at +4 °C for 15 min. The solution separated into distinct polar and nonpolar layers. Two extracts were prepared from each sample.

Preparation of Sample for HPLC

A 5 mL of upper hexane layer containing β-carotene was transferred to an amber coloured vial and evaporated to dryness under a stream of nitrogen at 40 °C (Caliper TurboVap LV, Hopkinton, MA). The residue was dissolved in 200 µL tetrahydrofuran (THF) containing 0.1 g/L buthylated hydroxytoluene (BHT) and diluted with 1800 µL methanol. The resulting extract was filtered through a 0.22-µm polytetrafluoroethylene (PTFE) filter (Sartorious AG, Goettingen, Germany) directly to an amber coloured auto sampler vial. The filtered extract was then immediately injected to HPLC without further delay.

Instrumentation and Chromatography

Separation and quantification of β-carotene were performed on a high performance liquid chromatography (Agilent 1200 series, Waldbronn, Germany) with a binary pump, a photo diode array (PDA) detector, a thermostatted auto-sampler, a degasser and a thermostatted column compartment. The chromatographic data were recorded and processed on Agilent 1200 series ChemStation rev.B.02.01 software. Isocratic separation was carried out on a C_{30} (5 µm) column (250x4.6 mm) (Phenomenex Inc., Los Angeles, CA) with a C_{30} (5 µm) guard column (10x4.0 mm) (Phenomenex Inc.). Mobile phase consisted of methanol: tert-buthylmethylether (65:35, v/v) solution containing 0.1 g/L BHT with a flow rate of 1.0 mL/min. Sample injection volume was 50 µL and column temperature was set at 30 °C. The detector was set at 450 nm, i.e., the absorption maximum of β-carotene in mobile phase. β-carotene in sulfited-DA samples was identified by comparing retention time and absorption spectra of unknown peaks with external reference standard (Sigma, St Louis, MO). Quantification of β-carotene was carried out using a calibration curve (R^2 = 0.994).

Surface Color Measurements

The surface color of sulfited-DAs was measured with a light reflectance spectrophotometer (Minolta CM-3600d, Osaka, Japan). The spectrophotometer had an 8 mm diameter viewing area. Measurements were recorded in L^* (lightness), $+a^*$ (redness) and $+b^*$ (yellowness) according to CIE (Commission Internationale de I'Eclairage) color coordinates, using the CIE C* illuminant which corresponds to the difference between average daylight and UV component. Saturation (Chroma, C^*) and color tone (hue angle, $h°$) were calculated from a^* and b^* color coordinates using the following equations:

$$C^* = (a^{*2} + b^{*2})^{1/2} \quad [1]$$

$$h° = \tan^{-1}(b^*/a^*) \quad [2]$$

Microbial Analyses

A representative sample (approximately 120 g) was taken from sulfited-DAs, and each apricot was aseptically cut into halves to obtain a uniform sample, and then a 30 g of subsample was aseptically transferred into a screw capped 500 mL flask. A portion of 90 mL sterile 0.1% (w/v) peptone water (PW) (Merck Co., Darmstad, Germany) was added just above the DA sample, and left for 15 min at ambient temperature (20 °C). By this way, the sample was slowly rehydrated, the recovery of injured cells was increased and the prevention of microorganisms from osmotic shock was carried out (17). Then, the remaining portion of PW was added to the rehydrated sample. The flasks were shaken vigorously for 1 min by a flask shaker (Griffin & George Ltd., UK) operating at half of the maximum speed corresponding to 1000 oscillation per min at ambient temperature. Further decimal dilutions were carried out in PW, and appropriate dilutions were later transferred onto appropriate media as follows.

Total mesophilic aerobic bacteria (TMAB) and

total psycrophilic aerobic bacteria (TPAB) were determined on plate count agar (PCA) with pour plate method. The plates were incubated for TMAB at 28 °C for 48 h and TPAB at 7 °C for 10 d. The enumeration of total yeasts and molds was carried out in yeast extract glucose chloramphenicol (YGC; Merck Co.) following an incubation period at 28 °C for 7 d. Meanwhile, for the enumeration of xerophilic molds in DA samples, dichloran glycerol agar (DG-18; Merck Co.) was used and the plates were incubated at 28 °C for 7 d. Lactic acid bacteria (LAB) were enumerated on MRS agar (De Man Rogosa Sharpe; Merck Co.) with pour plate method, overlayed with 5 mL of the same medium and incubated at 28 °C for 120 h. Total *Enterobacteriaceae* were enumerated in violet red bile dextrose agar (VRBDA; Merck Co.) with pour plate method, by overlaying with 5 mL of the same medium and incubated at 37 °C for 24 h. For the enumeration of *Staphylococcus* spp., the serial dilutions were spread on Baird-Parker agar (fortified with egg yolk and potassium tellurite) (BP; Merck Co.) and the plates were incubated at 37 °C for 48 h. All microbiologic analyses were done according to the APHA Compendium for the Microbiological examination of foods (18), and performed in replicate samples and dilutions of each replicate were plated in 2–4 replicate petri dishes.

Statistical Analyses

Experimental data were analyzed by using the Minitab statistical software, version 15 (Minitab Inc., State College, PA). SO_2 concentration was considered as the main effect. Statistical differences among means were determined by the Duncan's multiple range tests at the 5% significance level.

RESULTS and DISCUSSION

Moisture Content and Water Activity (a_w)

The moisture contents and a_w values of DAs containing SO_2 at different levels were presented in Table 2. There were differences in moisture contents and aw values of samples. The moisture contents and aw values of apricots ranged from 17.66 to 24.45% and 0.611 to 0.667, respectively. According to Codex standard for DA (19), the moisture content of DAs shall not exceed 25% when the preservatives such as SO_2 and/or sorbic acid are used. In this study, the levels of moisture in samples never reached to the maximum level specified in the Codex standard. Table 2 showed that there were no systematic differences between SO_2 concentrations and moisture contents, and aw values.

pH and Titratable Acidity

The pH and titratable acidity values of DA samples containing SO_2 at different levels were determined and the changes of these values with SO_2 levels were shown in Figure 1(A). As expected, there were significant differences in pH (3.98-5.28) and titratable acidity (0.72-2.53 g/100 g) of SDA samples. Excellent correlations were found between SO_2 level and pH (r = –0.913) and titratable acidity (r = 0.983). As the levels of SO_2 in DAs increased, the pH values decreased and the titratable acidity values increased (Figure 1(A)).

The increase in titratable acidity values can be attributed to the formation of sulfurous acid (H_2SO_3) in SDAs. As a result of reaction between SO_2 and water in apricots, H_2SO_3 forms (4). This, in turn, causes to increase in the titratable acidity

Table 2 Physical and microbiological properties of dried apricots containing SO_2 at different concentrations

Samples	Moisture (%)	Water activity	Reflectance colour values					TMAB (log cfu/g)
			L*	a*	b*	C*	h°	
0	17.94± 0.04G*	-	-	-	-	-	-	3.45± 0.72A
1	24.45± 0.02A	0.616± 0.001C	32.68± 4.82	6.34± 2.09	14.42± 3.97	15.79± 3.56	66.36± 3.00	1.99± 0.28C
2	18.86± 0.02F	0.616± 0.003C	30.09± 2.75	5.99± 2.02	13.07± 3.47	14.42± 3.27	65.23± 3.56	1.92± 0.04C
3	21.22± 0.00C	0.614± 0.001C	35.08± 3.45	6.34± 2.15	14.42± 2.98	15.79± 3.15	66.36± 4.00	2.14± 1.19BC
4	23.80± 0.30B	0.641± 0.001B	30.64± 2.16	5,99± 0.98	13.07± 3.48	14.42± 3.58	65.23± 3.04	1.95± 0.15C
5	17.66± 0.00G	0.611± 0.001C	31.30± 3.28	5.71± 1.20	15.30± 3.21	16.36± 2.80	69.29± 3.16	1.91± 1.10C
6	20.87± 0.13D	0.641± 0.001B	31.49± 5.24	5.37± 2.02	15.93± 3.35	16.84± 3.63	71.44± 4.39	2.26± 1.14B
7	19.41± 0.04C	0.667± 0.001A	33.78± 5.08	5.67± 2.56	14.98± 3.27	15.81± 2.99	70.75± 3.28	2.03± 1.10C

L*: Lightness; a*:Redness; b*: Yellowness; C*: Chroma; h°: Hue angle.
cfu: Colony forming unit; TMAB: Total mesophilic aerobic bacteria.
*Values are expressed in mean ± standard error.
A–G : Different letters in the same column are significantly different at 5 % level.

values of SDA samples [3]. This reaction is given below:

$$SO_2 + H_2O \rightarrow H_2SO_3 \quad [3]$$

In water, H_2SO_3 has two different dissociation steps by forming hydrogen ions according to the following two reactions [4, 5] (5) :

$$H_2SO_3 \rightarrow 2H^+ + SO_3^{2-} \quad [4]$$

$$H_2SO_3 \rightarrow HSO_3^- + H^+ \quad [5]$$

Hydrogen ions supplied by dissocation of H_2SO_3 also caused to decrease in pH values of SDA samples.

Browning Values

The effects of different SO_2 concentrations on the browning of DAs were shown in Figure 1(B). An excellent correlation was found between SO_2 concentrations and browning values (r = – 0.981). As expected, the brown color formation of SDAs increased as the SO_2 concentrations in DA samples decreased. The browning values of DA samples ranged from 0.35 (non-sulfited DAs) to 0.08 A_{420}/g dry weight. For the samples containing 188 and 452 mg SO_2/kg, the browning values were 0.28 and 0.25 A_{420}/g dry weight, respectively. Nury *et al.* (1960) defined the limit for acceptability of color of DAs as the absorbance values reach to a value of 0.30 at 440 nm (20). Therefore, right after drying, the SO_2 content as low as 188 mg per kg DAs is sufficient for an acceptable color value, if the DAs will not be stored. If they will be stored, DA samples containing low levels of SO_2 should be definitely stored at low temperatures (below 20°C) to prevent brown color formation during storage (21).

Differences in Surface Color

The CIE *L**, a*, b*, *C** and *h*° color values were used to track the differences in surface color of DAs containing SO_2 at different levels (Table 2). No systematic changes in *L**, a*, b*, *C** and *h*° were detected for the samples containing SO_2 at different levels. This was due to different moisture contents of samples (Table 2). This observation was in agreement with the study of Özkan *et al.* (22) who found that as the moisture content of DAs increased, the L* (lightness), b* (yellowness), C* and h° color values increased while the a* (redness) value decreased. Although the color of DA samples containing SO_2 at different levels cannot be differentiated by the reflectance spectrophotometer, the color differences were apparent visually.

Changes in β-carotene Content

The carotenoid contents of DA samples containing SO_2 at different levels and the HPLC separation of carotenoids from DAs were presented in Figure 1(C). Among the 2 carotenoid peaks detected, only β-carotene was identified. β-carotene was the major carotenoid of sulfited-DAs. Similar to our results, previous studies also showed that β-carotene was the major carotenoid in apricots followed by β-carotene and lycopene (23), with the β-carotene representing more than 60–70% of total carotenoids of apricots (23, 24).

Although the role of SO_2 in retarding the breakdown of β-carotene during drying and storage was well-known (25), to date, what level of SO_2 concentration in DAs was sufficient to prevent the breakdown of β-carotene during drying was not determined. In the present study, the β-carotene contents of DAs containing SO_2 at different levels ranged from 24.7 to 36.2 mg/100 g dry weight. Non-sulfited apricots had the lowest β-carotene content (24.7 mg/100 g dry weight). DAs containing 188 and 452 mg SO_2/kg had similar β-carotene contents and had approximately 20% lower β-carotene content than the apricots containing SO_2 at higher levels. Similarly, the β-carotene contents of DAs containing over 791 mg SO_2/kg were almost the same. These observations clearly showed that higher concentration of SO_2 more effectively protected carotenoids in DAs. Similar to our findings, Zhao and Chang (25) found dehydrated carrots containing 223 mg SO_2/kg (84 mg β-carotene/100 g) had approximately 26% lower β-carotene content than the apricots containing 1460 mg SO_2/kg (114 β-carotene mg/100 g). Because of the strong antioxidant activity, SO_2 is very effective in protecting carotenoids against oxidation during drying. The result from the present study revealed that minimum SO_2 concentration, which is necessary to keep the β-carotene contents of apricots during drying, was 791 mg/kg.

The high recoveries of 95.7, 91.2 and 89.8% were achieved when 26.752, 13.348 and 6.653 mg of β-carotene were added, respectively. Since the recoveries were mostly over 90%, the loss of β-carotene during extraction was minimal

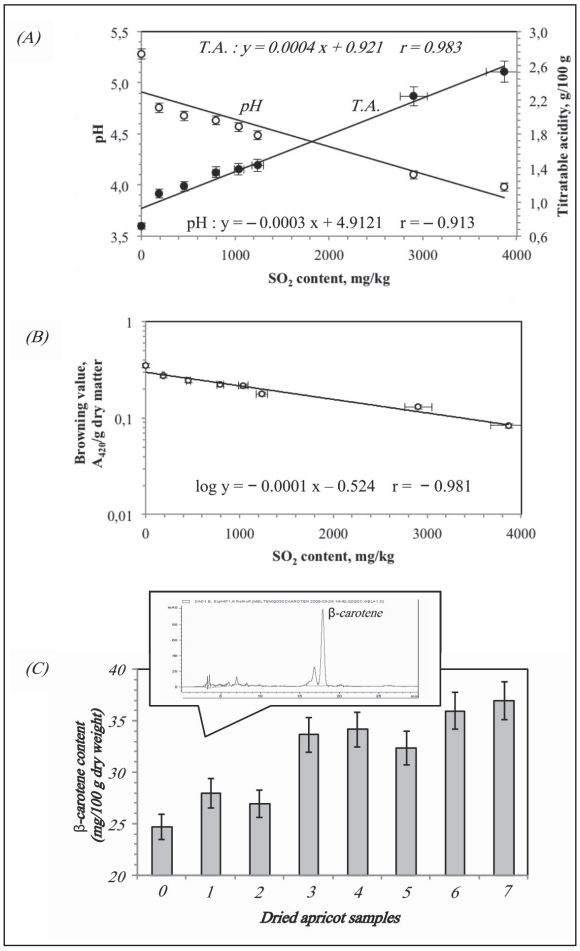

Figure 1. Effects of SO_2 concentrations on (A) pH and titratable acidity, (B) browning values and (C) β-carotene contents of dried apricots

(26). In other words, the high recoveries clearly indicated the accuracy of the carotenoid analysis.

Changes in Microbial Counts

The effects of different SO_2 concentrations on microbial counts (TMAB, TPAB, yeast and mold, xerophilic mold, LAB, *Staphylococcus* spp. and *Enterobacteriaceae*) in DAs were determined. As expected, the highest number of TMAB was found to be 3.45 log cfu/g in non-sulfited DAs. In general, fairly low numbers of microbial counts for total bacteria were detected. The lowest and highest number of TMAB was found to be 1.91 and 2.26 log cfu/g, respectively in SDA samples (Table 2). The results of microbiological analyses showed that there were insignificant (P>0.05) differences between the numbers of TMAB for the SDA samples with different SO_2 concentrations, except for the sample having SO_2 concentration of 2899 mg/kg which had slightly higher number of TMAB (P<0.05). Similar to the results of the present study, Sağırlı et al. (24) reported that high moisture DAs initially containing relatively low level of SO_2 showed a good microbial stability when stored at 5–30 °C for 8 months. They found as low as 98 mg SO_2 per kg of DAs containing 28.3% moisture after the storage at 30 °C for 8 months. TPAB, yeast and mold, xerophilic mold, LAB, *Staphylococcus* spp., and *Enterobacteriaceae* were below the lowest detection limit (<0.6 log cfu/g) in all the SDA samples.

The antimicrobial activity of SO_2 is well-documented (7). The most important reactions of SO_2 include with carbonyl groups such as aldehydes and ketones. As known, these groups are present in many critical biological molecules including proteins, enzymes, some enzyme cofactors (e.g. pyridoxal phosphate, folic acid) and sugars (27). Once SO_2 reacts with these molecules, these molecules can no longer participate the degradative reactions. SO_2 can also disrupt the disulfide bonds in protein structures, which provide maintenance of their tertiary structure, to form thiosulfonate and thiol pairs [6] (27):

$$–RSSR– + SO_3^{2-} \leftrightarrow –RSSO_3– + –RS– \quad [6]$$

In this way SO_2 may inhibit many of the critical enzymes within microbial cell (27).

CONCLUSION

DAs containing even very low level of SO_2 (188 mg/kg) showed excellent microbial stability right after drying. The growth of pathogenic microorganisms, such as *S. aureus* and Enterobacteriaceae, was not detected in all SDA samples. TMAB were very low and TPAB, yeasts and molds were below the lowest detection limit in all the SDA samples. The main color criteria for DAs are the formation of browning and oxidation of β-carotene. β-carotene was the major carotenoids in DAs. Both color criteria closely depend upon the SO_2 concentration which should be over 791 mg SO_2/kg to prevent the formation of brown color and the oxidation of β-carotene. Reflectance color values did not produce reproducible results due to the low moisture content of DAs which made color measurements very difficult.

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