Production of Tannins from *Acacia nilotica* Pods for the Leather Manufacturing Industry – Extractions, Characterization, and Optimization Using Design of Experiment

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The pods of *Acacia nilotica* were successfully utilized for the extraction of tannins using a methanol/water extraction medium. The experimental design employed for the extraction was a central composite design that enabled the evaluation of the effects of time, temperature, and methanol concentration on the dependent variables of extraction yield, total phenolic contents (TPC, as gallic acid equivalents (GAE)), and antioxidant activity (AA, as millimoles of ascorbic acid equivalents (AAE)). Response surface models were developed for the three responses, and statistical analysis of variance was performed to determine the optimum values of the independent variables and the coefficients of determination. The maximum extraction yield of 46.2 wt% (pod extract) was achieved at 200 min, 85 °C, and 40% methanol concentration. Similar conditions also led to the attainment of 50.7 wt% TPC and 51.3 mmol AAE/100 g pod. The coefficients of determinations were 0.9750 for extraction yield, 0.9626 for TPC, and 0.9774 for AA, which indicated that the model equations obtained fitted the experimental data. The result of the retanning of the leather using the extracted *A. nilotica* tannins also showed that the tear and tensile strength, as well as the elongation at break, of the leather samples were within the range obtained when chestnut extract and chrome tannin were used.

Keywords: Acacia nilotica pod; Extraction; Central composite design; Optimization

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INTRODUCTION

Briefly, *Acacia nilotica* is a plant that is 5 to 20 m high. It is thickly spherical in shape with its stems and branches blackly colored. It has pinkish-grey slashed and fissured bark, which exudes a reddish colored and low-quality gum. Its spines are straight, light, thin, and supplementary paired, usually between 3 and 12 and approximately 5 to 7.5 cm long in young trees, whereas the mature trees are commonly found without thorns. Its leaves are bipinnate and have up to 3 to 6 pairs of pinnulae with each pinnulae having 10 to 30 pairs of leaflets. It also contains seeds in pods. These *Acacia nilotica* pods are the versatile and dominant part of the plant that has been a source of organic compounds known as polyphenolics (Zhao *et al.* 2012).

Although the importance of this compound to the plant itself has not been well established in the literature, it is a phytochemical that contributes immensely to different

chemical groups, such as alkaloids, phenols, volatile essential oils, resins, steroids, oleosins, phenolics, glycosides, and most importantly tannins (Chaovanalikit and Wrolstad 2004). Aside from these phytochemicals, it has also been revealed that *A. nilotica* pods contain different profiles of bioactive components that include gallic acid, *m*-catechol, isoquercetin, glucopyranoside, and their derivatives, (+)-catechin-5-gallate, apigenin 6,8-C-di- glucoside and its derivatives (Melone *et al.* 2013), carotene, crude proteins, arabinose, calcium, and selenium (Rubanza *et al.* 2005).

Among all the phytochemicals stated above, tannins are the most extensively studied, especially as they are applied to the leather industry, and these are reported in recent articles (Covington 2011; Nasr *et al.* 2017). However, more research still needs to be done, especially in regards to its optimum production in mass scale as required in the leather industry. Tannins are generally regarded as chemicals that are rich in polyphenolics and have the capacity to form complexes with metal ions and proteins by a well-defined mechanism. Specifically, tannins are used for the clarification of antimicrobial activity in beverages as well as conversion of hides and skin into leather (Silvestre and Gandini 2008). Generally, leather tanning can be regarded as the conversion of proteins in hides and skins into leather by successive unit operations that include unhairing, degreasing, and desalting, as well as tanning (Covington 2004). Tannins used in the leather industry are classified as natural compounds and are divided into condensed and hydrolysable tannins. In this context, the emphasis of this study will focus on vegetable hydrolysable tannins extracted from *A. nilotica* pods. Generally, the essence of tanning is to make sure that the hide and skin are in stable condition and to prevent further putrefaction or decomposition after the tanning process. Tanning also provides successive cross-linkages between the collagen in the hide and skin, which ensures changes to its physical and chemical resistance (Thanikaivelan *et al.* 2005; Al-Hussaini and Mustapha 2016). The effect of the crosslinking is also to enhance the thermodynamic stability of the formed bond as a result of the changes to the macromolecules in the tanned hide and skin. It is important to state here that the tanning process also provides multiple hydrogen bonds and ionic interactions with the protein functional groups that are present in the hide and skin (Dixit *et al.* 2015; Pollach *et al.* 2016).

There is presently a renewed interest in the harvesting of vegetable tannins from different plant sources as an alternative to non-renewable mineral synthetic tannins, such as chromium-containing tannin products, due to the environmental concern of the latter in contributing to the pollution load from the leather industry (Dixit *et al.* 2015). Tannins, which is a class of phenolic compound that can be obtained from higher plants such as quebracho, mimosa, and chestnut, as well as the *A. nilotica* pod. These plants have been traditionally used for the tanning of leather (Kampa *et al.* 2003). It has been reported that traditionally tanned leather that used *A. nilotica* pods showed high resistance to microbial activity and putrefaction (Moure *et al.* 2001; Kampa *et al.* 2003; Okuda 2005; Babayi *et al.* 2011; Sugimoto *et al.* 2013; Deye *et al.* 2016). Other previous work on the extraction of tannins from *A. nilotica* species has been reported (Elgailani and Ishak 2014). These authors extracted tannins form different part of the acacia species and it was found that a mature pod has the highest percentage of tannins (22.2%) as compared to the bark and leaves (11.8% and 10.5%, respectively) of the same acacia species. Similarly, they also used hide powder methods to determine the tannin content of bark, leaves, and pods of the *A. nilotica*. Although this method has been in used for decades, it is time consuming and a lot reagents are required.

Generally, the application vegetable tannin extracts such as *A. nilotica* pod for leather tanning has several advantages over the synthetic chrome tannin. Some of the

advantages are retaining of the original outlook of the leather, eco-friendliness, and longer life, *etc*. The disadvantage of vegetable tanning is that the maturity of the *A. nilotica* pod as well as other weather conditions such as rainfall, soil fertility will affect the quality of the tannins (Arife *et al.* 2017). Hence, the use of modern methods of extraction of tannins from *A. nilotica* pods will play a vital role in the repositioning of the leather industry into greater efficiency of producing quality leather. To achieve this goal, the operating conditions for tannin extraction are an important parameter in obtaining the quality of extraction as well as the extraction efficiency. Some of the operating conditions necessary for optimizing the extraction efficiency in terms of quality and yield include temperature, time, and extraction solvent used, as well as the solid-to-liquid ratio used (Pinelo *et al.* 2005; Spigno *et al.* 2007; Chupin *et al.* 2013). It has been revealed in the literature that a methanol/water solvent mixture is the best extractive solvent for recovering total phenolic compounds from different plant species, such as from *Eucalyptus globulus* bark (Barakat *et al.* 2012; Kaushik *et al.* 2012; Asiedu *et al.* 2016). Apart from its good extractive potentials, it has also been reported that phenolic compounds extracted using this solvent have a strong resistance to antioxidants (Vázquez *et al.* 2008, 2009) when compared to single solvents or aqueous solutions such as sodium sulphate or sodium hydroxide (Chinsembu 2015).

EXPERIMENTAL

In this work, the methanol/water extractive medium were utilized. Apart from the careful selection of this extractive medium, the operating conditions of temperature, time, and methanol-to-water ratio were varied as independent variables to obtain the optimum extraction yield, total phenolic content (TPC), and antioxidant activity (AA) as the dependent variables. A central composite design was used for the optimization of the selected dependent variables through a reduced number of experiments. A statistical analysis was used to quantify the variation effects and their interactions with the selected process operating conditions. Response surface models for extraction yield, TPC, and AA of the tannins were developed, whereas an analysis of variance (ANOVA) was performed to determine the coefficient of determination (R^2) of these models. Part of the extract was used for the retanning of leather, which was originally tanned with chromium or chestnut extracts, to confirm the efficacies of the extracted tannins from *A. nilotica* pods.

Materials

Material collection and preparation

A. nilotica pods were collected from a forest reserve in the northern part of Nigeria. They were first air-dried in the absence of light to decrease their moisture content to approximately 20 wt%. The dried pods were then ground using a grinder (154GH; Putian Yangxin Machinery Company, Putian, China) to increase the surface area for solvent extraction.

Extraction of the tannin

The tannins were extracted using a 6-L M/K batch digester (M/K Systems, Inc., North Adams, MA, USA). The temperature and time as well as liquid recirculation ratio for different methanol/water solvents were determined for the ground pod after the extraction. The liquid-to-solid ratio was maintained at 4 L of the extraction medium/kg pods. The extraction reaction was batch-wise and the moisture contained in the pod was

measured, and it represented the amount of water used for the extraction. A constant heating rate of 3 °C/min was continually used during the heat-up period until the required temperature was reached; once this was achieved, the contact time was measured using a stopwatch. At the end of each extraction, the crude tannins extract was allowed to cool down to room temperature. Then, it was clarified after which nitrogen gas was bubbled into it and later kept in a refrigerator for subsequent characterization.

Statistical Experimental Design

The independent variables of time (X_1: 40, 200, or 360 min), temperature (X_2: 27, 85, or 150 °C), and methanol concentration (X_3: 0, 40, or 80%) were selected based on previous related work as described in the work of Pinto *et al*. (2013) (Table 2), and their influences on the selected dependent variables (Y_n) were studied using a full factorial central composite design (CCD) (*i.e*., 19 experiments) as designed by the Design Expert 11.0.0 software (Stat-Ease, Inc., Minneapolis, MN, USA). This software aided the experimental design, modeling, and optimization. It was also used to perform the statistical analysis of variance (ANOVA). The dimensionless coded values of the independent variables were obtained using Eq. 1; the dependent variables (Y_n) were the total extraction yield (Y_{EE}), TPC (Y_{TPC}), and AA (Y_{AA}) present in the extracted tannin. Equation 1 is as follows,

$$X_i = \frac{x_i - x_0}{\Delta x}, \text{ where } i = 1, 2, \ldots, k \quad (1)$$

X_i is the dimensionless coded value, x_0 is the value of x_i at the central point, and Δx is the step change (Alhaji *et al*. 2017).

Table 1. Ranges and Levels of the Experimental Design of the Independent Variables

Independent Variables	Factors (X_i)	Range and Level		
		-1	0	+1
Time (min)	X_1	40	200	360
Temperature (°C)	X_2	27	85	150
Methanol Concentration (%)	X_3	0	40	80

The required minimum, center, and maximum points were -1, 0, and +1, respectively. Each independent variable was incorporated into the CCD, which allowed for the design of the experiment and the construction of a second-order quadratic model (Eq. 2) for the dependent response variable, Y_n,

$$Y_n = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_{11}X_1^2 + b_{22}X_2^2 + b_{33}X_3^2 + b_{12}X_1X_2 + b_{13}X_1X_3 + b_{23}X_2X_3 \quad (2)$$

where Y_n represents the response variables for the extraction yield (wt%), TPC (wt% GAE on pod), or AA (mmol AAE/100 g Pod)**,** b_0 is the error, b_1, b_2, and b_3 are the coefficients for the linear effects, and b_{11}, b_{22}, and b_{33} are the quadratic coefficients, while b_{12}, b_{13}, and b_{23} are the coefficients for interaction effects (Alhaji *et al.* 2017*)*. Hence, the total number of experiments was 19 (2^3 [8] + 2*3 [6] + 5 = 19) and values in brackets represent the coded values of the independent variables (Table 2).

Table 2. Experimental Runs and Independent Variables (Actual and Coded Values) with Both Experiment and Predicted Values of the Responses

	Independent Variables (Coded Value in Parentheses)			Responses					
	X_1	X_2	X_3	Experimental			Predicted		
Run	Time (min)	Temp. (°C)	Methanol Conc. (%)	Extraction Yield (wt%)	TPC (wt% GAE on Pod)	AA (mmol AAE/100 g Pod)	Extraction Yield (wt%)	TPC (wt% GAE on Pod)	AA (mmol AAE/100 g Pod)
1	40 (-1)	27 (-1)	0 (-1)	6.7	7.2	5.2	8.7	7.1	7.1
2	360 (+1)	27 (-1)	0 (-1)	9.4	10.9	9.3	10.5	12.3	9.6
3	40 (-1)	150 (+1)	0 (-1)	14.6	9.5	11.7	14.6	12.1	10.9
4	360 (+1)	150 (+1)	0 (-1)	16.7	21.2	14.9	17.2	22.2	16.3
5	40 (-1)	27 (-1)	80 (+1)	20.8	24.3	21.8	20.3	23.3	19.9
6	360 (+1)	27 (-1)	80 (+1)	22.4	22.5	17.9	22.3	19.8	18.2
7	40 (-1)	150 (+1)	80 (+1)	25.7	29.4	24.5	24.8	28.3	23.9
8	360 (+1)	150 (+1)	80 (+1)	29.7	31.6	27.3	27.8	31.1	25.1
9	40 (-1)	85 (0)	40 (0)	33.4	32.9	29.3	32.7	31.4	30.7
10	360 (+1)	85 (0)	40 (0)	34.6	33.7	32.4	35.1	34.7	32.6
11	200 (0)	27 (-1)	40 (0)	35.3	31.5	35.9	32.5	32.6	35.2
12	200 (0)	150 (+1)	40 (0)	35.9	42.2	38.6	38.1	40.1	40.6
13	200 (0)	85 (0)	0 (-1)	36.7	46.9	41.2	33.1	41.1	38.5
14	200 (0)	85 (0)	80 (+1)	40.6	47.4	44.9	44.1	52.9	49.3
15	200 (0)	85 (0)	40 (0)	43.1	50.7	51.3	44.7	48.3	48.5
16	200 (0)	85 (0)	40 (0)	45.7	49.4	46.7	44.7	48.3	48.5
17	200 (0)	85 (0)	40 (0)	46.2	42.3	49.4	44.7	48.3	48.5
18	200 (0)	85 (0)	40 (0)	45.7	48.9	52.6	44.7	48.3	48.5
19	200 (0)	85 (0)	40 (0)	46.9	47.6	44.5	44.7	48.3	48.5

Methods

Total non-volatile solids

The extracted tannins were characterized according to the TAPPI T652m-89 (1989) standard to determine the amount of non-volatile solids. This method is a standard method fully described in the work of Song *et al.* (2010). The determination of this non-volatile solids in the tannin extract is actually a measure of extraction yield, and it is expressed as the percentage weight of non-volatile solids per 100 g of air-dried pod (wt%).

Determination of TPC

The TPC determination was quantified using the Folin-Ciocalteu method as explained by Neiva *et al.* (2015). This method involved the dilution of 0.5 mL of tannin extracts in aliquots, which were later kept in test tubes containing 1.5 mL of Folin-Ciocalteu reagent (95% pure, Sigma-Aldrich, St. Louis, MO, USA). Distilled water was used to make a solution up to 1:10 of the reagent, and 2 mL of $NaNO_3$ (> 99.9% pure, Merck, Kenilworth, NJ, USA) was added to it to make up 75 g/L of the solution. The resulting mixture obtained was allowed to stand for 5 min at 50 °C, and its UV-visible absorbance was measured with a spectrophotometer (UV-2501PC; Shimadzu, Mumbai, India) at 790 nm. A blank experiment was also prepared simultaneously using 0.5 mL of solvent. A calibration curve was produced using gallic acid (98% pure, Merck, Kenilworth, NJ, USA) with concentrations between 10 and 100 μg/mL. The results obtained were analyzed three times, and the mean value was calculated; TPC was expressed as grams of gallic acid equivalents (GAE) per 100 g of pods as a percentage (wt% GAE on pod).

Determination of extract antioxidant activity.

The AA activity of the extracted tannins was determined using an FRAP (ferric reducing antioxidant power) reagent (Feiya Chemical Industry, Jiangsu, China) as fully described by Song *et al.* (2010). The reagents used included 2,4,6-tri(2-pyridyl)-*s*-triazine (TPTZ) and iron chloride ($FeCl_3$), and both were analytical grade. The method was simple, reliable, and easily applicable. After the reduction of the Fe(III)-TPTZ complex to the Fe(II)-TPTZ complex, a blue color solution was obtained; its absorbance was determined by spectrophotometer (UV-2501PC; Shimadzu, Mumbai, India) measurement at 593 nm. An aliquot of diluted extract tannin, each 0.1 mL, was added to the FRAP reagent. All the experimental and blank samples were taken and tested using a calibration curve of L-ascorbic acid (98% pure, Allan Chemical Corporation, Ringwood, NJ, USA) standard solutions at 50 μmol/L under the same conditions. The analyses were conducted three times, and the mean value calculated and recorded against each experimental condition. The AA was expressed in millimoles of ascorbic acid per 100 g of pods (mmol AAE/100 g pod).

Retanning of the Leather Sample Using the Extracted Tannins from *A. nilotica* Pods

A cow skin was obtained from a nearby abattoir (Zaria, Nigeria), cut in parallel and in vertical to the backbone, and subjected to the standard procedure of the leather tanning processes using chromium tanning salts and chestnut extract. The leather pieces obtained were retanned using laboratory leather tanning drums (Z-LM201; HK Zion Industry, Guangdong, China). Four different assays were performed to compare the efficacy of the tannin extracts from *A. nilotica* pods with that of chestnut and synthetic chromium tannin extracts.

With all the assays performed on the leather, it was ensured that the water-to-skin ratio was maintained at 2, while the continuous addition of the synthetic chrome tannin was to ensure that final concentration was not more than 1 wt% (a represents leather sample). A total of 1% and 3% chrome extract were used for assay1 and assay2 and it contained 75% of tannins extract. In the last assay, *A. nilotica* tannin extracts were used for the retanning. These values are presented in Table 3, which depicts the extracted tannins contained in the *A. nilotica* pods per 100 g of leather as a weight percentage. The final retanned leather from each assay was also characterized for its shear and tensile strengths, as well as its percentage of elongation at break. The methods used in these characterizations were the ones used by the Society of Leather Technologists and Chemists (Font Vallès *et al*. 2010).

Table 3. Percentages of Tanning Agents Used in the Assays for the Retanning Process

	Wt% [a]		
Trial	**Chestnut Tannin Extracts**	***A. nilotica* Pod Tannin Extracts**	***Chromium Sulphate 33% basicity***
1	0	0	1
2	0.75	0	1
3	2.1	0	1
4	0	0.80	1

[a]Represents wet blue material known as leather

RESULTS AND DISCUSSION

The independent variables of time (X_1), temperature (X_2), and methanol-to-water ratio (X_3) as well as their effect on the three responses, Y_{EE}, Y_{TPC}, and Y_{AA}, of the *A. nilotica* pod extracts were examined. The 19 experimental runs are shown in Table 2. The experimental and predicted values of the responses are also shown as obtained from the CCD.

The experimental and predicted values agreed with one another. The optimization aimed at providing the set of values of independent variables that will meet the defined goals of maximizing tannin extraction yield, as well TPC and AA. The response surface methodology provided a clear picture of how these conditions affected all the three response variables.

Modeling and Optimization of Tannin Extract from *A. nilotica* Pod

The obtained experimental and predicted values are depicted in Table 2 for extraction yield, TPC, and AA. Figure 1 also shows that the data for each of the three responses were equitably spread close to one another in a linear fashion. Hence, this implied that the obtained plots showed a close agreement between the predicted model values and the experimental (Actual) values.

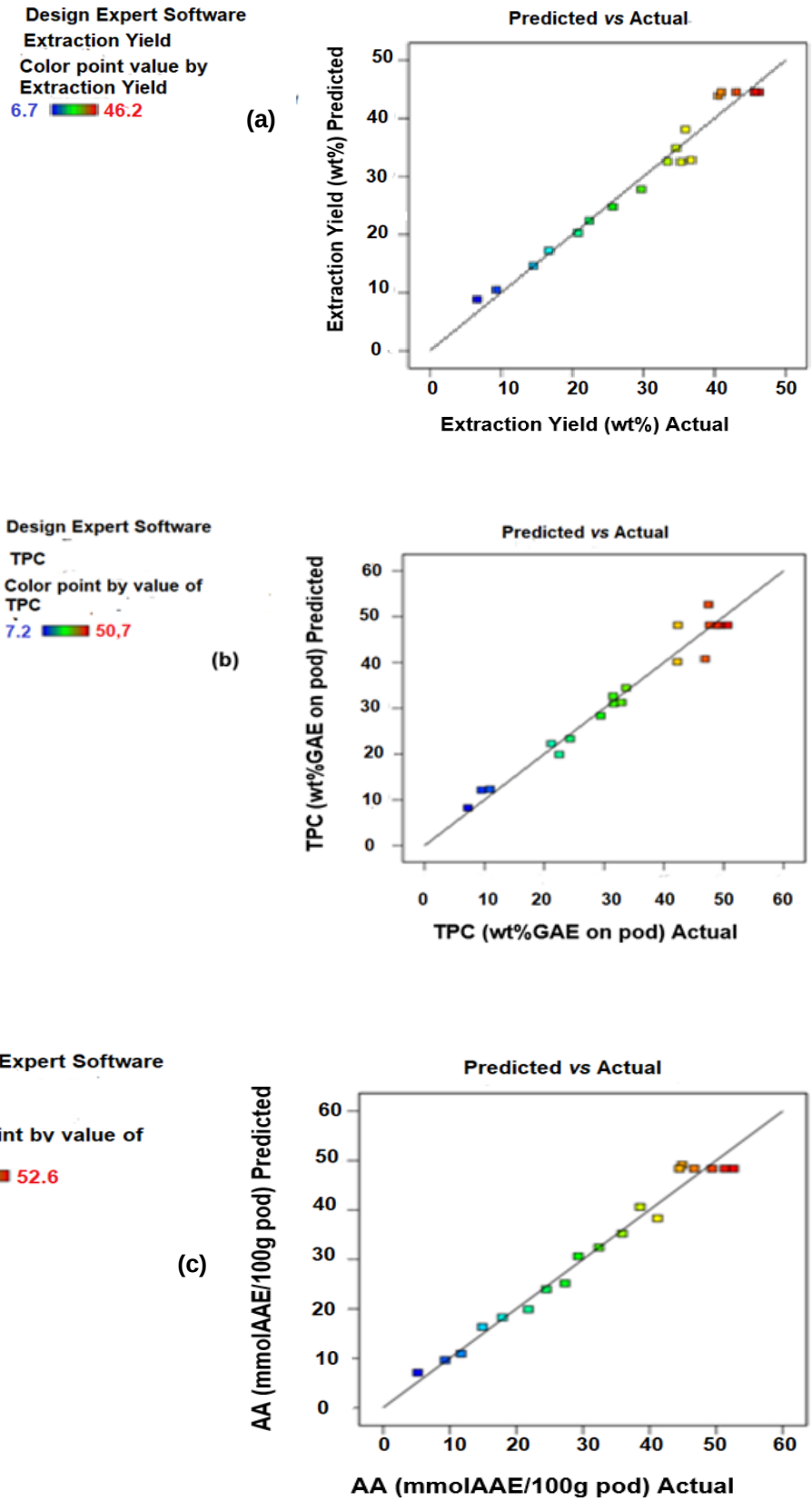

Fig. 1. Model prediction and actual values of the three response variables: (a) extraction yield, (b) total phenolic content, and (c) anti-oxidant activity

The model equations for the three responses in terms of coded values were obtained as shown in Eqns. 3 to 5:

$$Y_{\mathrm{EE}} = 44.77 + 1.16X_1 + 2.80X_2 + 5.5X_3 + 0.2328X_1X_2 + 0.100X_1X_3 - 0.3241X_2X_3 - 10.82 - 6.17X_3^2 \quad (3)$$

$$Y_{\mathrm{TPC}} = 48.39 + 1.68X_1 + 3.75X_2 + 5.95X_3 + 1.52X_1X_2 - 1.87X_1X_3 + 0.2809X_2X_3 - 11.99X_2^2 - 1.43X_3^2 \quad (4)$$

$$Y_{\mathrm{AA}} = 48.56 + 0.9381X_1 + 2.69X_2 + 5.41X_3 + 0.7157X_1X_2 - 1.05X_1X_3 + 0.050X_2X_3 - 16.85X_1^2 - 10.64X_2^2 - 4.65X_3^2 \quad (5)$$

Each of these equations consisted of linear, quadratic, and interaction effects of the independent variables on the various responses. The statistical significance of the mean square ratio was obtained using ANOVA. The three model equations obtained were verified at the 95% confidence level, and the values for the "lack of fit" (prob. > F) for extraction efficiency, TPC, and AA were 0.2674, 0.2971, and 0.5615, respectively (Table 3). All these values were greater than 0.05, which indicated that it was non-significant; hence, the proposed models fitted the experimental data (Table 4) (Alhaji *et al.* 2017). The adjusted R^2 values were 0.9500, 0.9252, and 0.9549 for extraction efficiency, TPC, and AA, respectively, and the values were close to the corresponding values for the R^2 (0.9750, 0.9626, and 0.9774, respectively); this indicated that the experimental data were reliably utilized in the determination of the response models. The adequate precision was also measured to indicate the signal-to-noise ratio, and it was found that the ratios for each of the three responses were greater than 4, which verified that the ratios obtain were desirable. Therefore, there was an adequate signal for the models to be used to traverse the design space.

Table 4. Statistical ANOVA for the Quadratic Models of the Three Responses

Independent Variables	Abbreviation	Extraction Yield	TPC	AA
Lack of Fit Prob. > F (p-value)	-	26.74%	29.71%	56.15%
Mean	Mean	30.74	33.16	31.55
Coefficient of Determination	R^2	0.9750	0.9626	0.9774
Adjusted R^2	Adj. R^2	0.9500	0.9252	0.9549
Coefficient of Variance	C.V.	9.12	11.78	10.17
Adequate Precision	A.P	17.541	15.675	18.0615

3D Response Surface Plots of the Responses

The response variables were assessed through the 3D plots as provided by the Design Expert statistical software. It provided useful information that revealed the diverse information on the interactive effects of the responses (Alhaji *et al.* 2017). It also showed the surface profile that helped in the precise and accurate determination of the optimum point.

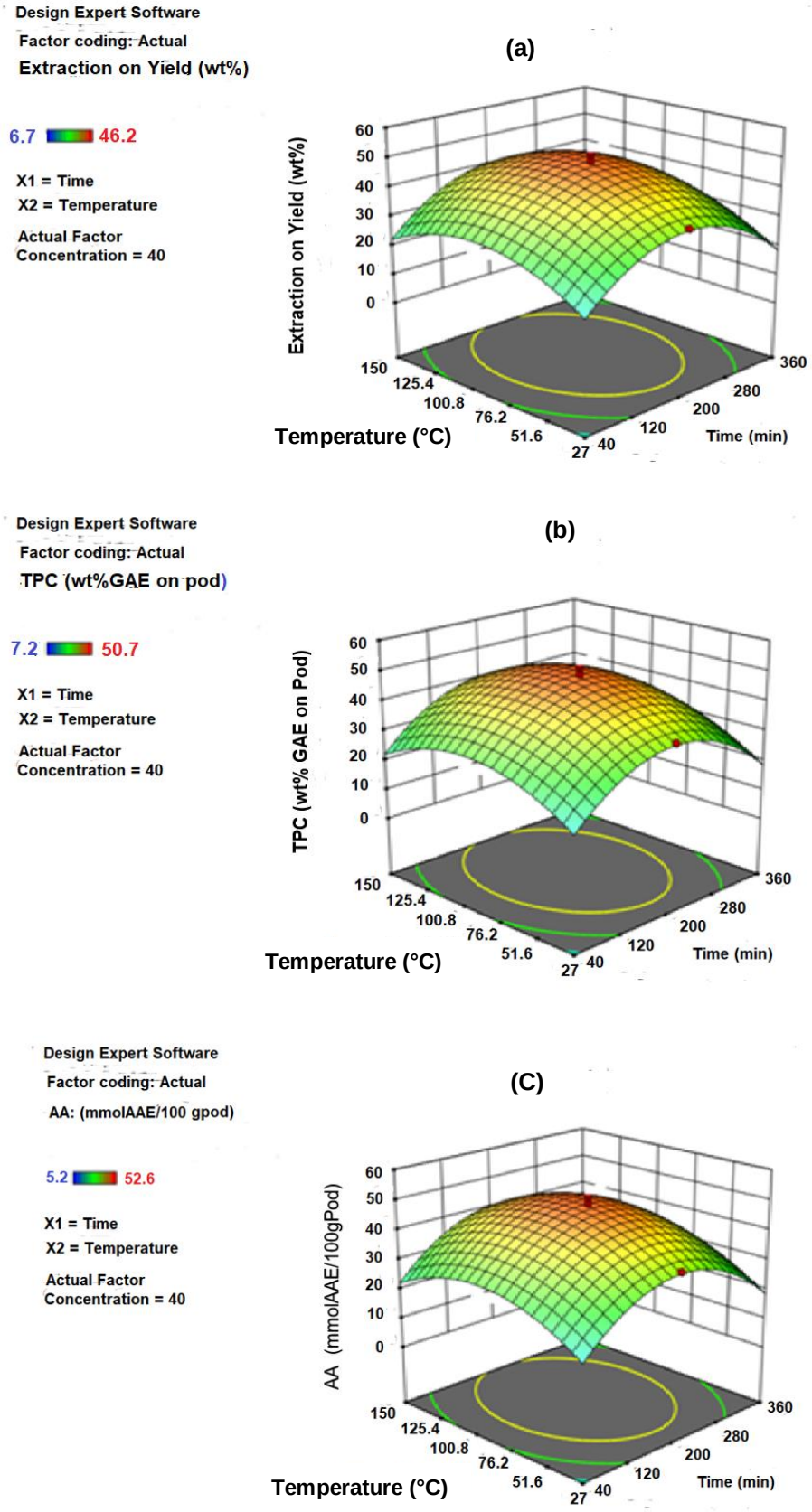

Fig. 2. 3D response surface of three response variables: extraction yield (a), TPC (b), and AA (c)

Figure 2 shows the surface plots with different peaks and values obtained from the model equations (Eqns. 3 to 5). As shown in Fig. 2(a), the methanol concentration was kept constant at 40%, while the temperature and time were varied to generate the extraction yield as a response variable. The maximum extraction yield was the highest (46.2 wt%) of the tested conditions at the boundaries (200 min, 85 °C, and 40% methanol). This value, when compared with the work of Vázquez *et al.* (2008) for the extraction *E. globulus* bark extract using NaOH, was higher. Under the same boundary conditions for extraction yield (Figs. 2(b) and 2(c)), the TPC and AA values were 50.7 wt% GAE on pod and 52.6 mmol AAE, respectively. Hence, it was deduced that the higher temperature favored the extraction yield of the tannins, while lower temperatures enhanced the TPC and AA contents and their selectivity. These assertions can be compared to the work of Spigno *et al*. (2007), in which the TPC and AA contents of the grape extracts were higher at the lower temperature. Similarly, at a constant methanol concentration, all the responses (except for the extraction yield) trended downward, which implied that time and temperature played a vital role in the overall interaction effects of the variables on the responses. Furthermore, the methodology used in this work was effective in understanding the peculiarity and composition of each of the independent variables on the responses studied.

Determination of the Tear Strength, Tensile Strength, and Elongation at Break of the Retanned Leather

As mentioned in the previous section, the essence of the retanning is to test the efficacy of the *A. nilotica* pod tannin extracts. This was achieved with the determination of the tear and tensile strength, as well as the elongation at break of the treated leather. After the subjection of the leather samples to these three physical tests, the results were plotted (Fig. 3).

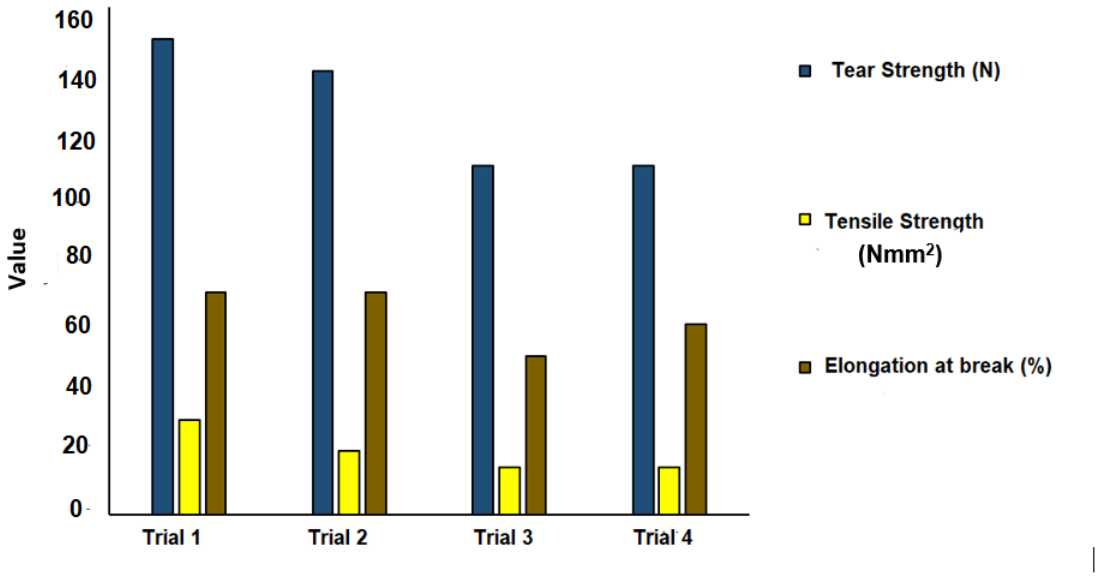

Fig. 3. Results of physical tests obtained after the retanning of leather with chromium tannin (Trial 1), chestnut extracts at 1% (Trial 2), chestnut extracts at 3% (Trial 3), and *A. nilotica* tannin extracts (Trial 4)

At the starting point, the concentrations of the chestnut extracts were kept at three different values of 0, 1, and 3 wt% for the three different trials (Trial 1, Trial 2, and Trial

3, respectively). It was ensured that the chestnut extract tanning content was kept at 75%; hence, the tannin concentration in Trial 2 and Trial 3 was at 0.75 and 2.7 wt%, respectively. Under these conditions, it was observed that all three physical test values obtained from *A. nilotica* pod tannin extracts were within those obtained for the chestnut extracts. It must be emphasized that the shear stress resistance of the retanned leather depends on the extraction concentration and extraction causticity. The causticity of the tannin extracts is related to the attainment of lower mechanical resistance, as well as lower elongation at break (Pinto *et al*. 2013). The roundness and fullness of the leather also depended on the causticity of the tannin extracts. In general, vegetable tannins, such as those extracted from *A. nilotica* pods or *E. globulus* bark, provide a high environmental resistance when used in the tanning of leather (Pinto *et al*. 2013). The *A. nilotica* plant is abundantly available and several million tons of its pods can be used for the production of tannins. Though the extracted tannin has been used to retan a sample leather and it was of good mechanical strength, it is important to further analyze the efficacy of the tannin using other parameters such as putrefaction rate and antibacterial activity of the retanned leather.

CONCLUSIONS

1 The central composite design (CCD) of experimentation, as incorporated in the Design Expert 11.0.0 statistical software, was successfully utilized to optimize three independent extraction conditions for the production of tannins from *A. nilotica* pods.

2 The optimal set of conditions, *i.e*., 200 min, 85 °C, and 40% methanol concentration obtained a maximum 46.2 wt% extraction yield, 50.7 wt% TPC, and 51.3 mmol AAE antioxidant activity.

3 This work further demonstrated the capacity of using *A. nilotica* pods for the extraction of tannins required in the leather industry.

ACKNOWLEDGMENTS

Special thanks are given to the Directorate of Research and Development of the Nigerian Institute of Leather and Science Technology (Zaria, Kaduna State, Nigeria) for supporting this research (Grant No. F01/NILEST/1256/2019/07).

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Article submitted: December 2, 2019; Peer review completed: January 24, 2020; Revised version received: January 31, 2020; Accepted: February 2, 2020; Published: February 6, 2020.
DOI: 10.15376/biores.15.2.2212-2226