## Fat Absorption Capacity of Alfalfa Hay Harvested at Three Stages of Maturity with Different Particle Sizes and Some Feedstuffs on Common Oil Sources

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Abstract Absorbance of extracted oil during oilseeds extrusion is important to produce high quality product. To date there is little information about fat absorption capacity (FAC) of common feedstuffs as absorbents in the extrusion process of oilseeds. The objective of current study was to determine the FAC of alfalfa hay harvested at three stages of maturity (SM; early bud, late bud and early flower) and with different particle sizes (2, 0.75 and 0.2 mm) and some feedstuffs as absorbent materials during oilseeds extrusion. In a completely randomized design, the FAC of experimental feedstuffs was different (P<0.05), and alfalfa hay had highest FAC among all absorbents (P<0.05). The FAC of alfalfa hay was not affected when particle sizes and oil sources changed (P>0.05). With advancing the SM of alfalfa hay, the FAC increased (P<0.05). Different combination of alfalfa hay and pistachio by-products had different FAC, and with an increase in the alfalfa hay portion, FAC increased (P<0.05). In conclusion, the FAC of alfalfa hay changed with advancing SM but not by different particle sizes or oil sources and different feedstuffs had various FAC.

Keywords: Absorbent, extrusion, oil seed, pistachio by-products.

### Introduction

Fat absorption capacity (FAC) of a material is one of numerous quality parameters that contribute to the overall acceptability of the material as an ingredient in prepared foods (Lin *et al.*, 1974). In producing extruded materials, particularly oilseeds, FAC evaluation is an important test for absorbents as it ascertains the propensity of the absorbent to absorb oil. The FAC test normally is used as the basis for selection of the absorbent materials to be mixed with

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oilseeds during the extrusion process (Eggie, 2010). Indeed, during the process of extrusion, several operations take place, such as grinding, hydration, mixing, shearing, thermal treatment, gelatinization, protein denaturation, destruction of microorganisms and some toxic compounds, shaping, expanding, and partial dehydration (Riaz *et al.*, 2005).

There are some inherent problems in the extrusion process of an oilseedbased feed ingredient. Under intense pressure in extrusion process, oil will be extracted from the seeds, and the recovery of this oil means that, it can be transfer at a later stage, to the animal feed. Furthermore, when this oil is extracted, it is exposed to high temperatures and air, which may probably increase the rate of oxidation and reduces the desirability of the feed ingredient from a farmer/producer perspective as it smells bad and tastes rancid. Excess oil that is not retained by the feed ingredient may be lost during transport and storage, which can be considered an economic loss, as the desired and health beneficial ingredient of oilseed is its oil (Eggie, 2010).

One suggested way to reduce oil losses during extrusion process, is to use different feed-approved absorbents (Akraim *et al.*, 2007; Eggie, 2010).

A large number of materials may have potential as absorbents in the extrusion process. The criteria for selecting absorbent is mainly based on the FAC of the absorbent and the cost of including that in the final extruded feed ingredient. One absorbent with the high FAC may cause high oil retention in final products. Oil retention is the ability of the extruded products to retain oil that it is related to the type of absorbent and the ratio of oilseed to absorbent in the extrusion process (Eggie, 2010). In the study of Eggie (2010) on extruded flax-based feed ingredients, alfalfa had higher FAC than soy hulls and corn gluten, probably as a result of its higher fiber content.

Another problem for extruded oilseeds with a high content of polyunsaturated fatty acids, such as linseed and sunflower seed, is the oxidation of these FAsduring the extrusion process and storage period (Frank *et al.*, 2002).

In some cases, while absorbents would have a good FAC, they are also high in natural antioxidants. Different studies reported that the polysaccharide (Liu *et al.*, 2010; Liu *et al.*, 2011) and peptide (Xie *et al.*, 2008) content of alfalfa have antioxidant effects in which with its higher fiber content and acceptable FAC (Eggie, 2010), may probably be a good absorbent for producing extruded oilseed products. Stage of maturity at harvest is an important factor affecting alfalfa hay fiber and probably its antioxidant content (Yari *et al.*, 2012).

To date there is a little information about the FAC of alfalfa hay at three stage of maturity (early bud, late but, and early flower), alfalfa hay with different particle size (as a physically characteristic) and some common feedstuffs. The primary objective of current study was to find FAC of alfalfa hay at different stage of maturity and particle sizes on different oil sources. The secondary objective was to find the FAC of common feedstuffs and compare them with alfalfa hay and alfalfa hay to pistachio by product whit different ratio as absorbents in the oilseed extrusion.

#### Materials and methods

#### Fat Absorption Capacity evaluation

At first, common feedstuffs (Table 1) were ground to pass through a 2mm screen. For assessing FAC the method of Lin *et al.* (1974) used. A 15 ml centrifuge tube was weighed and its mass recorded. Next, 0.3 g of absorbent material was weighed in a centrifuge tube. To the centrifuge tube, 3 ml of linseed oil was added. The tube and its contents were vortexed appropriately and centrifuged at 3,000 rpm for 30 minutes at room temperature. After centrifuging, the supernatant fluid was removed from the tube via pipette and the final mass of the tube and contents was recorded. This procedure was performed in triplicate for each material. The calculation used to determine FAC was:

%FAC= ((sample mass+absorbed oil mass)  $\div$  sample mass)  $\times 100$ 

Alfalfa hay samples used in current study was previously analyzed for chemical composition and nutrient availability and supply by Yari*et al.* (2012). To determine the effect of particle size of alfalfa hay on FAC, samples were ground to pass three sizes screen of 2, 0.75, and 0.2 mm. Different sources of oils were used in which two vegetable oils (linseed oil and soybean oil) were utilized as vegetable oil sources with different unsaturated fatty acid composition and one animal oil (butter) as an oil with saturated fatty acids. For FAC evaluation, the butter was first melted over medium heat.

Absorbent Materials					
Alfalfa hay (AL)	Citrus pulp (CP)				
Alfalfa hay (AL)	Barley grain (BG)				
Sugar beet pulp (SBP)	Limestone (LS)				
Wheat bran (WB)	Corn grain (CG)				
Soybean meal (SBM)	Sodium bicarbonate (SB)				

Table 1. Absorbent materials for Fat Absorption Capacity experiment

The second step of the FAC analysis assessed whether the most absorbent, "expensive" absorbents improve the FAC more than the less absorbent, "cheap"

absorbents. The most absorbent material was combined with a selection of less absorbent materials in the following ratios of 5:95, 10:90, and 50:50 (high: low) (Eggie, 2010). Given the high prices of alfalfa hay in Iran, this study utilized 5 different proportions (by mass) of alfalfa: pistachio by-products (66.66:33.33; 50:50; 33.33:66.66; 90:10 and 80:20) for FAC evaluation. The FAC of these blends was then tested using the methodology described by Lin *et al.* (1974).

#### Statistical analysis

Data of FAC for different absorbents, alfalfa hay with different stage of maturity, and FAC for a mix of alfalfa and pistachio by-products with different ratios were analyzed in a completely randomized design using the general linear model (GLM) procedure of SAS 9.2 (2003).

The statistical model used, was:

 $\mathbf{Y}_{ij} = \mathbf{\mu} + \mathbf{T}_i + \mathbf{e}_{ij}$ 

where  $Y_{ijk}$  is the observation;  $\mu$  is the overall mean;  $T_i$  is the effect of treatments; and  $e_{ij}$  is the random error.

Data on FAC for alfalfa hay with different particle sizes using different oils were analyzed using the (GLM) procedure of SAS 9.2 (2003) in a completely randomized design with a  $3\times3$  factorial arrangement with the following model:

 $Y_{ijk} = \mu + A_i + B_j + A_i \times B_j + e_{ijk}$ 

where  $Y_{ijk}$  is the observation of the dependent variable,  $\mu$  is the overall mean,  $A_i$  is the fixed effect of oils (*i*=3, linseed oil, soybean oil, and butter);  $B_j$  is the fixed effect of particle size (*j*=3, 2, 0.2 and 0.75 mm);  $A_i \times B_j$  is the fixed effect of the interaction between factor oil at level *i* and the factor particle size at level *j*; and  $e_{ijk}$  is the random error associated with the  $k^{th}$  replicate in cell (*i*, *j*). Significant differences between individual means were identified using Duncan's multiple range test at a (0.05) probability level.

#### **Results and discussion**

The stage of maturity had a significant effect on the FAC quality of alfalfa hay (P<0.05), as with advancing stage of maturity, the FAC quality of alfalfa increased linearly (Table 2).

 Table 2. Fat absorption capacity (% FAC) of alfalfa hay cut at three stages of maturity

 Stage of maturity
 SEM<sup>1</sup>

Brage of maturity				<b>DL</b> IN <b>I</b>
	Early bud	Late bud	Early flower	

FAC (%)	401.66 <sup>c</sup>	432.5 <sup>b</sup>	460.99 <sup>a</sup>	4.683
<sup>1</sup> SEM, standard err	or of mean; means	whit different supe	rscripts (a, b, c) are si	ignificantly

different (P<0.05).

Yariet al. (2012) reported that with advancing maturity alfalfa stem portion, fiber content increased and protein decreased. There were no interaction effects between oil source and particle size for the FAC parameter of alfalfa hay (Table 3). Different oil sources and the particle size of alfalfa hay had no effects on the FAC quality of alfalfa hay (P>0.05). While in current study the particle size of alfalfa hay had no impact on FAC, therefore, the chemical composition of alfalfa hay, especially fiber content, which changed with advancing maturity, probably is more related to FAC.

The FAC of data, exhibited there were significantly different among the materials (Table 4) probably as a result of different chemical composition (Eggie, 2010). Alfalfa hay had highest FAC compared to other absorbents (P<0.05). After alfalfa hay, wheat bran significantly had the highest FAC in contrast to other materials (P<0.05). In addition, the FAC of sugar beet pulp, citrus pulp, pistachio by-products, soybean meal, barley grain, and corn grain was at a medium level. The FAC quality of sodium bicarbonate and limestone was at a low level.

Eggie (2010) determined the FAC of several different materials and reported that Solka-Floc<sup>®</sup> and Arbocel<sup>®</sup>, pure cellulose products, had considerably higher FAC than the other absorbents (wheat bran, alfalfa, soy hulls, pea fiber, corn gluten, wheat gluten, beet pulp, corn, and bentonite). It seems that materials with higher NDF content have a higher tendency to absorb oil.

With increasing alfalfa portion in the mixtures of alfalfa hay: pistachio by-products, FAC increased (P<0.05), and the mixture with the higher content of alfalfa (alfalfa: pistachio by-products, 90:10), had the highest FAC quality as compared to other mixtures (Table 5). This result indicates that pistachio by-products has different potential of FAC and could not be totally substituted for alfalfa hay.In addition, it can be resulted that, combining a high performance material with a lower performance material would increase the effective FAC of the lower performance material (Eggie, 2010).

**Table 3.** Effect of different oil sources and particle size on fat absorptioncapacity (% FAC) of alfalfa hay

Oil source	SEM	Particle size	SE	P-value
	1		Μ	

Ite	Linsee	Soybea	Butter		2mm	0.75m	0.2m		Oil	Particl
m	d	n				m	m		sourc	e size
									e	
FA	298.5	289.22	289.1	9.029	298.4	292.58	287.5	9.02	0.708	0.701
С	4		6		3		8	9		
(%)										
$^{1}$ S	EM stan	dard arror	of means							

SEM, standard error of means

<b>Table 4.</b> Fat absorption capacity of different materials with linseed oil											
	Absorbent Materials <sup>1</sup>									SEM <sup>2</sup>	
Item	AL	WB	SBP	CP	PIS	SBM	BG	CG	SB	LS	SEM
%FA C	259.62 <sup>a</sup>	247.16 <sup>b</sup>	222.33 <sup>bc</sup>	212.99 <sup>c</sup>	211.16 <sup>c</sup>	198.83 <sup>cd</sup>	197.49 <sup>cde</sup>	<sup>e</sup> 188.16 <sup>de</sup>	179.5 <sup>e</sup>	145.83 <sup>f</sup>	9.612

<sup>1</sup>AL, Alfalfa hay; WB, Wheat bran; SBP, Sugar beet pulp; CP, Citrus pulp; PIS, Pistachio by-products; SBM, Soybean meal; BG, Barley grain; CG, Corn grain; SB, Sodium bicarbonate and LS, Limestone

<sup>2</sup>SEM, standard error of mean; means whit different superscripts (a, b, c) are significantly different (P < 0.05)

**Table 5.** Fat absorption capacity (%FAC) of mixed alfalfa and pistachio by-products with different ratio.

		Alfalfa: Pistachio by-products ratio						
Item	90:10	80:20	66:33	50:50	33:66			
FAC (%)	294.83 <sup>a</sup>	282.58 <sup>b</sup>	271.53 <sup>c</sup>	265.87 <sup>d</sup>	235.15 <sup>e</sup>	1.441		

<sup>1</sup>SEM, standard error of mean; means whit different superscripts (a, b, c) are significantly different (P<0.05).

#### Conclusion

The FAC of alfalfa hay increased with advancing maturity from early bud to early flower and different particle size of 2, 0.75, and 0.2 mm of alfalfa hay had no effect on its FAC. The FAC of alfalfa hay did not differ when oil sources changed. Common feedstuffs, had different FAC and pistachio by-products has comparable FAC with alfalfa hay.

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