

**UTILIZATION OF SOME NON-CONVENTIONAL
DIETS IN GROWING RABBITS FEEDING**

By

REHAB ABDEL-HAY MOHAMMED ABDEL-HAY

B.Sc. Agric. Sc. (Poultry Production), Ain Shams University, (2003)

M.Sc. Agric. Sc. (Poultry Nutrition), Ain Shams University, (2009)

This thesis submitted in partial fulfillment

Of

The requirement for the degree of

DOCTOR PHILOSOPHY

in

Agricultural Sciences

(Poultry Nutrition)

Department of Poultry Production

Ain Shams University

Faculty of Agriculture

2017

Approval sheet

**UTILIZATION OF SOME NON-CONVENTIONAL
DIETS IN GROWING RABBITS FEEDING**

By

REHAB ABDEL-HAY MOHAMMED ABDEL-HAY

B.Sc. Agric. Sc. (Poultry Production), Ain Shams University, (2003)

M.Sc. Agric. Sc. (Poultry Nutrition), Ain Shams University, (2009)

This thesis for Ph.D. degree has been approved by:

Dr. Mamdouh Omar Abd-Elsamee
Professor of Poultry Nutrition, Faculty of Agriculture, Cairo
University

Dr. Alaa El-Dien Abdel-Salam Hemid
Professor Emeritus of Poultry Nutrition, Faculty of Agriculture, Ain
Shams University

Dr. Sayed Ahmed Abdel-Fattah
Professor of Poultry Physiology, Faculty of Agriculture, Ain Shams
University

Dr. Fathy Abdel-Azeem Mohamed
Professor of Poultry Nutrition, Faculty of Agriculture, Ain Shams
University

Date of Examination: 15/ 2/ 2017

UTILIZATION OF SOME NON-CONVENTIONAL DIETS IN GROWING RABBITS FEEDING

By

REHAB ABDEL-HAY MOHAMMED ABDEL-HAY

B.Sc. Agric. Sc. (Poultry Production), Ain Shams University, (2003)

M.Sc. Agric. Sc. (Poultry Nutrition), Ain Shams University, (2009)

Under the supervision of:

Dr. Fathy Abdel-Azeem Mohamed Ahmed

Professor of Poultry Nutrition, Department of Poultry Production,
Faculty of Agriculture, Ain Shams University (Principal supervisor).

Dr. Sayed Ahmed Abdel-Fattah Mohamed

Professor of Poultry Physiology, Poultry Production Department,
Faculty of Agriculture, Ain Shams University.

Dr. Mohamed Helmy Mohamed Yacout

Head of Research, By-products Utilization Department, Animal
Production Research Institute, Agriculture Research Center.

ABSTRACT

Rehab Abd El-Hay Mohamed Abd El-Hay: Utilization of some Non-Conventional Diets in Growing Rabbits Feeding, Unpublished Ph.D. thesis, Ain Shams University, Faculty of Agriculture, Department of Poultry Production, 2017.

This experiment was conducted to study the reduction tannins and phenolics content (anti-nutritional factors) in *Calendula Officinalis* by-products (COP) using biological treatment. Clover hay was replaced by biologically treated (COP) at level 15 and 30% in growing New Zealand White rabbit diets. Effects on growth performance, nutrients digestibility, carcass traits, blood constituents, caecum traits and economic efficiency were studied. 84 unsexed weaned New Zealand White rabbits, six weeks old with an average live body weight from 850 to 900 g, were randomly assigned to 7 groups (12 rabbits each). Each group was divided into 4 replicates, (three rabbits each). The result showed that degradation of tannins and phenolics compound was more efficient when *Tricoderma reesi* were incubated with COP at 30°C. Data showed that rabbits fed diets containing 30% COP with *T. reesie* had the best live body weight, body weight gain, feed conversion, digestion coefficient, nutrient value, and the best weight of empty carcass and dressing percentage, and high economic efficiency followed by groups fed on treated roughage with poly ethylene glycol (PEG), at 14 weeks of age, when compared with control or other groups. It can be concluded that, the feasibility of using biologically treated COP to replace clover hay; it can be used as a feed supplement in the diet of rabbits it proved to be as a good replace of clover hay without any adverse effect on rabbit performance.

ACKNOWLEDGMENTS

First of all, thanks to the most generous God who helps me to finish this study

I wish to state my genuine gratitude and profound appreciation to prof. **Dr Fathy Abdel-Azeem Mohamed**, Professor of poultry Nutrition, Department of Poultry Production, Faculty of Agriculture, Ain Shams University, for his dependability in suggesting the topic, continuous leadership and guidance during the preparation and writing of this manuscript.

My honest thanks and sincere gratefulness are due to Prof **Dr. Sayed Ahmed Abdel-Fattah Mohamed**, Professor of Poultry physiology, Poultry Production department, Faculty of Agriculture, Ain Shams University, for his continuous and close supervision, and his unlimited effort in revising the thesis, preparation and writing of this manuscript are greatly appreciated.

Kind acknowledgment is also due to **Prof. Dr. Mohamed Helmy Mohamed Yacout**, Head of Research, By-products Utilization Department, Animal Production Research Institute, Agriculture Research Center for his support during the theoretical and practical work and for generous facilities and efforts offered to complete this study.

Also, I like to express my deepest appreciation and gratitude to **Dr. Ayman Abd El-Mohsen**, Head of Research, Nubaria Animal Production Research Station, Agriculture Research Center, for his diligent labor, encouragement and great help during the experimental work.

Deeply gratitude to Animal Production Department, Central Administration for Agriculture By-products Utilization Department, Animal Production Research Institute, Agriculture Research Center for generous facilities offered during the practical course. Deeply thanks to

all the staff member of Poultry Production Department, Faculty of Agriculture, Ain Shams University, for their help

Last but not least, great recognition are extended to my mother, my family for their fortitude and back-up through the progress of this work

CONTENTS

Item	Page
LIST OF CONTENTS	i
LIST OF TABLES	iv
LIST OF FIGURS	v
LIST OF ABBREVIATIONS	vi
1-INTRODUCTION	1
2-REVIEW OF LITERATURE	3
2.1. Scientific Classification	3
2.2. Morphological of <i>Calendula Officinalis</i> plants.	4
2.3. Different compounds of <i>Calendula Officinalis</i> .	5
2.4. Chemical parameters of <i>Calendula Officinalis</i> .	8
2.5. Traditional uses of <i>Calendula Officinalis</i> .	9
2.5.1. Effects of <i>C.Officinalis</i> on antimicrobial and antifungal activities.	10
2.5.2. Antioxidant effects.	12
2.5.3. Using herbal plants in livestock's nutrition.	14
2.6. Effect of <i>Calendula Officinalis</i> on blood parameters.	14
2.7. Natural toxins (anti nutritional factors).	17
2.7.1. Tannins.	18
2.7.1.1. Types of tannins.	19
2.7.2. Phenolic compound.	19
2.8. Effect of anti-nutritional factors on livestock's nutrition.	20
2.9. Role of dietary fiber in rabbits' nutrition.	24
2.10. Effect of fiber on growth rate and the digestibility of Nutrients.	25
2.11. Microflora in rabbits.	27
2.12. Improving the nutritive value of roughages.	29
2.13. Biological treatment.	30
2.14. Methods used for degradation toxic compound in plant substrates.	30
2.14.1 Using fungi for reducing anti-nutritional factors levels.	30
2.14.2. Effect of fungi and bacteria on degradation of tannins.	31
2.15. Effect of fungi and bacteria on chemical composition of crop residues.	32
2.16. Using polyethylene glycol (PEG) for reducing anti nutritional factors.	33
2.17. Effect of feeding animals on biologically treated agricultural by-products.	36
2.17.1. Animal growth performance.	36
2.17.2. Digestibility and nutritive value.	39
2.17.3. Volatile fatty acids in rabbits.	41
2.17.4. Carcass characteristics.	43

2.17.5. Blood constituents.	45
3-MATERIALS AND METHODS	46
3.1. Collecting of <i>Calendula Officinalis</i> by-product (COP) and Treatments.	46
3.2. Biological treatment.	46
3.2.1. Microorganisms.	46
3.2.2. Fungal Inoculum Preparation.	46
3.2.3. Scaling up of fungal biomass.	47
3.2.4. Cultivation procedures.	47
3.3. Treatment with Polyethylene glycol (PEG).	47
3.4. Treatments of <i>Calendula Officinalis</i> by-products (COP).	48
3.4.1. Untreated Diet.	48
3.5. Animals Experimental Design and Management.	48
3.6. Digestibility trials.	48
3.7. Collected Data.	51
3.7.1. Anti Nutritional Factors.	52
3.7.2. Instrument used.	52
3.7.3. Preparation of extracts.	52
3.7.4. Determination of total phenolic content.	52
3.7.5. Determination of tannin Content.	53
3.8. Live Body weight (LBW).	53
3.9. Body Weight Gain (BWG).	53
3.10. Feed Consumption (FC).	54
3.11. Feed Conversion Ratio (FCR).	54
3.12. Mortality Rate % (MR).	54
3.13. Slaughter Traits.	54
3.14. Carcass composition.	55
3.15. Caecum activity.	55
3.15.1. Determination of cecum total volatile fatty acid.	55
3.16. Blood Plasma analysis.	56
3.16.1. Determination of Anti-oxidant enzymes activities	56
3.16.1.1. Catalase (CAT) enzyme activity	56
3.16.1.2. Superoxide Dismutase (SOD)	57
3.16.1.3. Glutathione (GSH):	57
3.16.1.4. Glutathione peroxidase (GSHpx)	57
3.17. Determination of Plasma Total Protein (PTP):	57
3.18. Determination of Plasma Albumin	57
3.19. Determination of Plasma Globulin	58
3.20. Determination of liver function enzymes	58
3.20.1. Transaminases (AST) and (ALT)	58
3.21. Determination of Creatinine	58
3.22. Determination of Urea Nitrogen	58

3.23. Detemination of Total lipid	59
3.24. Detemination of Triglycerides	59
3.25. Detemination of Total cholesterol	59
3.25.1. High Density Lipoprotein cholesterol (HDL).	59
3.25.2. Low Density Lipoprotein Cholesterol (LDL).	59
3.26. Economic efficiency.	59
3.27. Statistical analysis.	60
4. RESULTS & DISCUSSION	61
4.1. Chemical analysis of treated and untreated.	61
4.2. Rabbit performance.	64
4.2.1. Live body weight.	64
4.2.2. Daily weight gain.	65
4.2.3. Feed consumption.	66
4.2.4. Feed conversion ratio (FCR).	68
4.3. Effect of treated and untreated Calendula Officinalis by-products on nutrients digestibility.	71
4.4. Effect of treated and untreated Calendula Officinalis by-products on nutritive value.	76
4.5. Effect of untreated and treated Calendula Officinalis by-products levels on nitrogen utilization.	77
4.6. Effect of treated and untreated Calendula Officinalis by-products on carcass characteristics.	80
4.7. Effect of treated and untreated Calendula Officinalis by-products on carcasses meat chemical analysis of growing rabbits.	83
4.8. Effect of treated and untreated Calendula Officinalis by-products level on some blood hematological and biochemical traits of growing rabbits.	87
4.9. Effect of untreated and treated Calendula Officinalis by-products level on antioxidant enzymes activity.	91
4.10. Effect of treated and untreated Calendula Officinalis by-products level on caecum activities.	95
5. SUMMARY	97
6. REFERENCES	101
ARABIC SUMMARY	

LIST OF TABLES

No.	Title	Page
1	Composition of the experimental diets and their chemical analyses.	50
2	Chemical analysis (%) of treated and untreated Calendula Officinalis by-products.	63
3	Effect of Calendula Officinalis by-products treated or untreated on performance of New Zealand rabbits.	70
4	Effect of Calendula Officinalis by-products treated or untreated on nutrient digestibility of New Zealand rabbits.	75
5	Effect of Calendula Officinalis by-products treated or untreated on carcasses characteristics of New Zealand rabbits.	79
6	Effect of Calendula Officinalis by-products treated or untreated on chemical composition of carcasses meat of New Zealand rabbits.	82
7	Effect of Calendula Officinalis by-products treated or untreated on some blood traits of New Zealand rabbits.	86
8	Effect of Calendula Officinalis by-products treated or untreated on antioxidant enzymes activities of New Zealand rabbits.	90
9	Effect of Calendula Officinalis by-products treated or untreated on caecum characteristics of New Zealand rabbits.	94
10	Effect of calendula officinals by-products treated or untreated on economic efficiency of New Zealand rabbits.	96

LIST OF ABBREVIATIONS

CO	Calendula Officinalis
COP	Calendula Officinalis by-products
PEG	Polyethylene glycol
TR	Tricoderma reesi
DM	Dry Matter
OM	Organic Mater
CP	Crud Protein
CF	Crud Fiber
EE	Ether Extract
NFE	Nitrogen Free Extract
DE	Digestible energy
TT	Total Tannins
TP	Total Phenolics
BWD	Body Weight Gain
FC	Feed Consumption
FCR	Feed Conversion Ratio
MR	Mortality Rate
VFA	volatile fatty acid
PTP	Plasma Total Protein
AST	Aspartate aminotransferase
ALT	Alanine aminotransferase
HDL	High Density Lipoprotein cholesterol
LDL	Low Density Lipoprotein Cholesterol
VLDL	Very low density lipoprotein cholesterol
R.E.EF	Revenue economic efficiency
ADF	Acid detergent fiber
MF	Marigold flower
HU	Haugh unit

BF	Butanolic fraction
CM	Calendula meal
CT	Condensed tannins
FTS	Fungal treated straw
UTS	Urea treated straw
DMD	Dry matter digestible
CFD	Crud fiber digestibility
TDN	Total digestible nutrient
NI	Nitrogen intake
UN	Urinary nitrogen
FN	Feces nitrogen
ND	Nitrogen digestible
NB	Nitrogen balance
N	Nitrogen
CPD	Crud protein digestible
DCP	Digestible crud protein
GAE/g	Gallic acid equivalent / gram of dry matter
SCP	Single cell protein
CAT	Catalase enzyme activity
SOD	Superoxide dismutase
GSH-Px	Glutathione peroxidase
GSH	Glutathione
MDA	Malondialdehyde
EM	Effective microorganism
FDA	Food and Drug Administration
RE/g	reagent equivalent/ gram
UV	Ultra violet

INTRODUCTION

In Egypt, the shortage in feedstuffs is considered the main problem facing the development of animal production. Mean time, there are vast quantities of residues of crops, vegetables and fruits and herbs, such as potato vines, eggplant shoots, pea vines, watermelon leaves and tomato vines, artichoke by-products, calendula officinalis that are wasted and not used in animal feed.

Herbal plants by-products are the cheapest source of essential amino acids, vitamins and mineral. It is mainly used because of its various biological activities. But, the presence of inherent toxic factors or anti-nutritional components in certain plants is considered one of the major obstacles in harnessing their full benefits and nutritional value (**Lewis and Fenwik, 1987**). *Calendula officinalis* L. (English marigold, pot marigold) belongs to the Asteraceae (Compositae) family; is an annual herbaceous plant, native of Mediterranean countries (**Danielski et al., 2007**). *C. officinalis* can be broadly applied as an antiseptic, anti-inflammatory and cicatrizing as well as a light antibacterial and antiviral agent (**Khalid et al., 2010**). The plant has been reported to contain mainly polyphenols such as *p*-hydroxybenzoic, salicylic, vanillic, caffeic, gallic acids (**Gora et al., 1979; Gong et al., 2012**), acylated flavonoid-*O*-glycosides and methoxylated flavonoids, amino acids (**Abasova et al., 1995**), alkaloids, carotenoids, saponins, tannins (**Duke, 1992**), high molecular weight polysaccharides (**Wagner et al., 1984**) and triterpenoid monoesters (**Neukirch et al., 2004**). Alpha-cardinol (**Chalchat et al., 1991**), delta-cadinol, delta-cadinine and gamma murolene (**Marczal et al., 1987**) have been identified in the essential oil. Previous studies showed that different species of this plant, as well as different cultivars of the same species, were markedly different in the content of their phenolics, flavonoids and antioxidant activities as well as antioxidant

INTRODUCTION

properties were in correlation with the content of total phenolics and flavonoids (**Ercetin *et al.*, 2012**).

Therefore, methods like as physical, chemical, physicochemical and biological treatments are essential for improving nutritive value of such by-products. Some disadvantages of these methods, such as nutritional loss, reduced sensory quality, and high cost of needed equipments, have limited their practical use in animal feed (**Gowda *et al.*, 2007**).

Polyethylene glycol (PEG) is a polymer that binds tannins irreversibly over a wide range of pH, and reduces the formation of protein-tannin complex (**Jones and Mangan, 1977**). However, the biological treatments were used to reduce anti-nutritional factor by using certain fungi, such as *Aspergillus parasiticus*, in degrading aflatoxins, possibly through fungal peroxidases. Fermentation with yeast has also been found effective in destroying patulin and rubratoxin B (**Lopez-Garcia and Park, 1998**). Also, **Oda *et al.* (2002)** reported that filamentous fungi, which have been isolated from potato sprouts, are able to produce an enzyme which degraded ability to glycoalkaloids.

The aim of this study was:

- 1- To investigate the possibility of reducing phenolics and tannins anti-nutritional factors in herbal plants residues (such as *calendula officinalis*) by using biological and chemical treatments.
- 2- To investigate the possibility of replacing clover hay by biologically and chemically treated herbal plants residues (*calendula officinalis*) for up to 15% or 30%, and their effects on productive performance of growing rabbits.

REVIEW OF LITERATURE

2.1. Scientific Classification

Kingdom:	Plantae
Unranked:	Angiosperms
Unranked:	Eudicots
Unranked:	Asterids
Order:	Asterales
Family:	Asteraceae
Tribe:	Calenduleae
Genus:	Calendula
Species:	<i>Calendula officinalis</i>

Calendula officinalis is a short-lived aromatic **herbaceous perennial**, growing to 80 cm, with sparsely branched lax or erect stems. The leaves are 5–17 cm, hairy on both sides, and with margins entire or occasionally waved or weakly toothed. The **inflorescences** are yellow, comprising a thick **capitulum** or flower head 4–7 cm diameter surrounded by two rows of hairy bracts; in the wild plant they have a single ring of ray florets surrounding the central disc florets. The disc florets are tubular and **hermaphrodite**, and generally of a more intense orange-yellow color than the female, tridentate, peripheral ray florets. The flowers may appear all year long where conditions are suitable. *Calendula officinalis* is widely cultivated and can be grown easily in sunny locations in most kinds of soils. Although perennial, it is commonly treated as an **annual**, particularly in colder regions where its winter survival is poor and in hot summer locations where it also does not survive.