Saudi Journal of Biological Sciences 26 (2019) 821–827

Contents lists available at ScienceDirect

Saudi Journal of Biological Sciences

journal homepage: www.sciencedirect.com

Original article

Effects of dodder total flavone on polycystic ovary syndrome rat models induced by DHEA combined HCG

Mingsan Miao *, Mengfan Peng, Zhengwang Zhu, Xiaoli Yan, Zhenzhen Wei, Mengyan Li

College of Pharmacy, Henan University of Chinese Medicine, Zhengzhou 450000, China

ARTICLE INFO

Article history:
Received 15 August 2018
Revised 19 February 2019
Accepted 19 February 2019
Available online 19 February 2019

Keywords:
PCOS
Pathological changes
Dodder total flavone
Biochemical indicators

ABSTRACT

Aims: Explore the effects of dodder total flavone on polycystic ovary syndrome (PCOS) rat models induced by dehydroepiandrosterone (DHEA) combined human chorionic gonadotropin (HCG).
Methods: Except the blank group, the rest of the rats were injected with DHEA 6 mg/100 g on the back of the neck and 1.5 IU HCG each day, for 21 consecutive days. On the 16th day of modeling, vaginal smear was performed to select the model rats, which were randomly divided into model group, dacin-35 group, large, middle and small dose dodder total flavonoids groups, and given the medicine for three weeks. At the end of the last administration, take samples, so as to calculate the ovaries and uterus indexes, measure serum LH/FSH ratio, P, PRL and INS levels, fixed the uterus and pancreas in 10% formalin solution and stained with HE to observe the morphological changes of the organs. And measure the expression of TNF-α and IGF-l proteins in ovaries by immunohistochemistry.
Results: Compared with the blank group, ovarian and uterine indexes, serum LH/FSH ratio, serum PRL and INS levels, ovary TNF-α and IGF-l protein expression were significantly increased, and significant pathological changes were observed in the uterine and pancreatic tissues in model group ($P < 0.01$). While the serum P level decreased significantly ($P < 0.01$), Compared with the model group, the ovarian and uterine indexes, serum LH/FSH ratio, serum P, PRL and INS levels, ovary TNF-α protein expression were significantly decreased in large, middle and small dose dodder total flavonoids groups ($P < 0.01$); The expression of IGF-1 protein was decreased and uterus pathological changes were improved in different extents ($P < 0.01$ or $P < 0.05$), pancreas pathological changes were improved significantly ($P < 0.01$).
Conclusion: PCOS rat models was successfully replicated. Dodder total flavone can protect PCOS rats induced by DHEA combined HCG by different action pathways.

1. Introduction

Polycystic ovary syndrome (PCOS) is a reproductive endocrine metabolic disorder characterized by insulin resistance accompanied by persistent ovulation disorder and hyperandrogenemia (Elbohoty Ahmed et al., 2016). Western medicine believes that genetic factors, high luteinizing hormone, glycolipid metabolism disorder (Daghestani, 2018), hyperinsulinemia and insulin resistance (Giandalia), hyperandrogenemia (Li et al., 2018), low degree inflammatory reaction (Jamilian), obesity (Rezvan et al., 2018), abnormal ovarian function and hyperpuberty are the etiology and pathogenesis of PCOS (Leng and Wei, 2018; Zuo and Fu, 2018). PCOS belongs to the category of "late menstruation" and "amenorrhea" in traditional Chinese medicine (TCM). In TCM, ovum is called "reproductive sperm", TCM believes that kidney storage sperm, leading to menstrual production. Deficiency of kidney leads to deficiency of qi, blood and liver qi stagnation. In turn, it leads to spleen apoplexy, which leads to different TCM syndromes of PCOS. Based on clinical data mining, kidney deficiency ranks first among the TCM syndromes of PCOS, and is often accompanied by blood deficiency, blood stasis, liver depression and other syndromes (Huang et al., 2018). TCM emphasizes the combination of dialectic and disease differentiation. For the root cause of PCOS disease is kidney deficiency, it is necessary to use Chinese medicine to supplement kidney for intervention. Dodder is a kind of traditional Chinese medicine commonly used in the treatment of PCOS. Studies have shown that its main chemical component flavonoids

* Corresponding author.
E-mail address: miaomingsan@163.com (M. Miao).
Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

https://doi.org/10.1016/j.sjbs.2019.02.007
1319-562X/

can have a multidirectional effect on the endocrine function of hypothalamic-pituitary-gonads, such as improving the level of estrogen in the body, protecting spermatogenic cells of male rats, and improving glycolipid metabolism (Zhao et al., 2018; Ren et al., 2018; Huang et al., 2016). However, there is no relevant research report on the intervention of total flavonoids from dodder in PCOS. Exogenous increase of DHEA level can create a high androgen state in patients with polycystic ovary syndrome, resulting in decreased secretion of follicle-stimulating hormone in the body, follicular development blocked, follicular atresia and lack of ovulation (Liu et al., 2018). HCG can play the role of LH, and can simulate the high LH environment in patients with polycystic ovary syndrome (PCOS) due to gonadal axis disorder, aggravating the PCOS secretion disorder. Therefore, making the PCOS rats induced by DHEA combined with HCG as the research object, to observe the effect of total flavonoids of dodder on PCOS rats.

2. Experimental materials

2.1. Drugs and reagents

Dodder total flavone, Nanjing zelang biotechnology co. LTD, the total flavonoid content of dodder was 51.25% by UV detection. Diane-35 (Ethinylestradiol and Cyproterone Acetate Tablets), Schering GmbHCo. Produktions KG, batch no:333A; Dehydroepiandrosterone (DHEA), Aladdin, batch no: B1227012; Human chorionic gonadotropin (HCG), Hangzhou animal medicine factory, batch no: 161008, Veterinary drug approval number: veterinary drug (2010) 110201281.

Anti-GnRH, Anti-GnRH-R and Anti-TNF-α polyclonal antibody, Beijing boorson biotechnology co. LTD, batch no: AC12243558S, AF11177594S and AG06274 154S; Anti-AR polyclonal antibodies, Abcam. Batch number: GR310012-4; Peroxidase labeled sheep rabbit-IgG ii-antibody, Wuhan bode bio-engineering co. LTD, batch no: BST12E27C55; Detection kits for LH, FSH, P, PRL and INS ELISA in rats, Suzhou Calvin biotechnology co. LTD, batch no: 20170623SR. Anti-IGF-1 polyclonal antibody, Wuhan bode bio-engineering co. LTD, Batch no: BST10E21C 54.

2.2. Experimental animals

SD rats, SPF level, male, 23 days old, 70 subjects. Jinan Pengyue experimental animal breeding co, LTD. License no: SCXK (lu) 20150005; Animal certificate no: 37009200006387; Training certificate no: 1612; Animal facilities use certificate no: 00013517.

2.3. Experimental apparatus

BX61 electric microscope, OLYMPUS, Japan; AR1140/C electronic analytical balance, Ohaos (Shanghai) co. LTD; Type 680 enzyme marker, Bio-rad company, USA; KDC-160HR high speed refrigerating centrifuge, Hkust innovation LTD; TGL-168, high-speed table centrifuge, Shanghai anting science instrument factory; HWS12 electric thermostat water bath, Shanghai yiheng scientific instruments co. LTD.

3. Experimental method

3.1. Moldeling and drug delivery (Miao et al., 2018)

Ten rats were randomly selected as the blank group, the rest of the rats were injected with DHEA 6 mg/100 g on the back of the neck and 1.5 IU HCG each day, for 21 consecutive days. On the 16th day of modeling, vaginal smear was performed to select the model rats. Rats with successful modeling were selected and randomly divided into model group, dacin-35 group (0.3392 $mg{\cdot}kg^{-1}$), large, middle and small dose dodder total flavonoids groups (200 $mg{\cdot}kg^{-1}$, 100 $mg{\cdot}kg^{-1}$, 50 $mg{\cdot}kg^{-1}$), 10 in each group, and given the medicine for three weeks. The blank group and the model group were given the same amount of solvent.

3.2. Test indicators and methods

Two hours after the last administration, draw blood from abdominal aorta, and serum was separated to measureLH/FSH ratio, P, PRL and INS. Ovaries and uterus were weighed so as to calculate viscera index. Uterus and pancreas were fixed in 10% formalin solution for HE staining and observe morphological changes. The aprotein expression of TNF-α and IGF-l in the ovary was measured by immunohistochemistry.

4. Statistical methods

SPSS21.0 statistical software was used to statistically process the experimental results. The measurement data are expressed as mean value ± standard deviation ($\bar{x} \pm s$). Single factor analysis of variance was used for inter-group comparison. The homogeneity of variance was tested by LSD method, and the heterogeneity of variance was tested by Games-Howell method. The hierarchical data is validated with Ridit.

5. Experimental results

5.1. Effect of dodder total flavone on ovary index and uterus index of PCOS rat models induced by DHEA combined HCG

As shown in Table 1: Compared with the blank group, the ovary index and uterus index of the model group were significantly increased ($P < 0.01$). Compared with the model group, dacin-35 group, large, middle and small dose dodder total flavonoids groups could significantly reduce the ovary index and uterus index of PCOS rats ($P < 0.01$).

5.2. Effect of dodder total flavone on serum LH/FSH ratio of PCOS rat models induced by DHEA combined HCG

As shown in Table 2: Compared with the blank group, the serum LH/FSH ratiol in the model group increased significantly ($P < 0.01$). Compared with the model group, the serum LH/FSH ratio in the dacin-35 group, large, middle and small dose dodder total flavonoids groups decreased significantly ($P < 0.01$).

5.3. Effect of dodder total flavone on serum P, PRL and INS levels of PCOS rat models induced by DHEA combined HCG

As shown in Table 3: Compared with the blank group, PRL and INS levels in the serum of the model group increased significantly ($P < 0.01$), serum P level decreased significantly ($P < 0.01$). Compared with the model group, the serum PRL and INS levels decreased significantly ($P < 0.01$), P level increased significantly in ($P < 0.01$).

5.4. Effect of dodder total flavone on TNF-α and IGF-l protein expression in ovariary of PCOS rat models induced by DHEA combined HCG

As shown in Table 4: Compared with the blank group, the mean optical density of TNF-α and IGF-l protein in ovary of the model group increased significantly ($P < 0.01$). Compared with the model group, the mean optical density of TNF-α and IGF-l in ovary was

Table 1
Effect of dodder total flavone on ovary index and uterus index of PCOS rat models induced by DHEA combined HCG.

Group	*n*	Dose (mg/kg)	Uterus index (mg/g)	Ovary index (mg/g)
Blank group	10	—	1.08 ± 0.17	0.45 ± 0.06
Model group	10	—	2.35 ± 0.27$^{\Delta\Delta}$	0.56 ± 0.08$^{\Delta\Delta}$
Dacin-35 group	10	0.339	1.04 ± 0.21**	0.43 ± 0.07**
Dodder total flavone	10	200	1.05 ± 0.15**	0.45 ± 0.07**
	10	100	1.05 ± 0.07**	0.42 ± 0.07**
	10	50	1.05 ± 0.10**	0.47 ± 0.07**

Note: Compared to the blank group, $^{\Delta}$represents $P < 0.05$, $^{\Delta\Delta}$represents $P < 0.01$; Compared to the model group, *represents $P < 0.05$, **represents $P < 0.01$.

Table 2
Effect of dodder total flavone on serum LH/FSH ratio of PCOS rat models induced by DHEA combined HCG.

Group	n	Dose (mg/kg)	LH/FSH
Blank group	10	—	3.55 ± 0.28
Model group	10	—	4.68 ± 0.66$^{\Delta\Delta}$
Dacin-35 group	10	0.339	3.61 ± 0.28**
Dodder total flavone	10	200	3.64 ± 0.33**
	10	100	3.80 ± 0.39**
	10	50	3.94 ± 0.54**

Note: Compared to the blank group, $^{\Delta}$represents $P < 0.05$, $^{\Delta\Delta}$represents $P < 0.01$; Compared to the model group, *represents $P < 0.05$, **represents $P < 0.01$.

significantly decreased in the dacin-35 group, large, middle dose dodder total flavonoids groups ($P < 0.01$), small dose dodder total flavonoids group could significantly reduce the mean optical density of TNF-α in ovary ($P < 0.01$) and obviously reduce the mean optical density of IGF-l ($P < 0.01$).

5.5. Effect of dodder total flavone on uterine tissue of PCOS rat models induced by DHEA combined HCG

Pathological observation of uterine tissue in rats (see Fig. 1): In the blank group, endometrial glands were normal, papillary arrangement, covered by a single layer of columnar epithelium. The nuclei were arranged in a neat arrangement, and no nuclear consolidation or nuclear fragmentation was observed. In the model group, endometrial glandular hyperplasia, with microscopic pseudoplastic columnar epithelium. At magnification, the nuclei are prominent but mostly fragmented, and inflammatory cells such as neutrophils can be seen in the field of view. In the dacin-35 group, endometrial glands were irregular or tubular arrangement, covered by a single layer of columnar epithelium, nuclei arranged neatly. In the large and middle dose dodder total flavonoids group, the glands were normal, papillary arrangement, and no inflammatory cells were seen. In the small dose dodder total flavonoids group, endometrial gland was slightly hyperplasia, and the glands are mostly papillary and tubular arrangement. The nuclei were arranged in a neat order, but the nucleoli were not obvious. A small number of inflammatory cells can be seen in the stroma below the horizon.

The uterine tissues of each group were measured according to the semi-quantitative standard, and the results were shown in Table 5.

As shown in Table 5: By Ridit test, the uterine endometrium of rats in the blank group was normal. Compared with the blank group, the endometrial glands showed significant hyperplasia and inflammatory infiltration in the model group ($P < 0.01$). Compared with the model group, the pathological changes of uterus in rats was significantly improved in the dacin-35 group, large, middle dose dodder total flavonoids groups ($P < 0.01$), small dose dodder total flavonoids group could obviously improve the pathological changes of uterus in rats ($P < 0.05$).

5.6. Effect of dodder total flavone on islet tissue of PCOS rat models induced by DHEA combined HCG

Pathological observation of islet tissue in rats (see Fig. 2): In the blank group, the islet shape and structure of the rat pancreas were normal, and the cells were arranged in a compact and orderly manner. In the model group, abnormal lesions appeared in pancreatic islet tissue. It suggests that patients with polycystic ovary syndrome may have islet tissue lesions, and islet lesions may also promote the occurrence of polycystic ovary syndrome through relevant mechanisms, aggravating the patient's condition. Under the light microscope, it is obvious that the islet volume of PCOS model group is larger than that of the blank group. However, the islet volume of each group which were given drugs decreased significantly. The pancreas of each group of animals was selected to measure 10 pancreas islets, and the mean value of the widest and narrowest part of the pancreas was obtained by measuring microscale, then the diameter of the pancreas islet was obtained, and the measured results were shown in Table 6.

The uterine tissues of each group were measured according to the semi-quantitative standard, and the results were shown in Table 5.

As shown in Table 5: Compared with the blank group, the pancreatic islet diameter in the model group increased significantly ($P < 0.01$). Compared with the model group, dacin-35 group, large, middle and small dose dodder total flavonoids groups could significantly reduced the diameter of pancreatic islets ($P < 0.01$).

Table 3
Effect of dodder total flavone on serum P, PRL and INS levels of PCOS rat models induced by DHEA combined HCG ($\bar{x}$±s, n = 10).

Group	n	Dose (mg/kg)	P (ng/mL)	PRL (ng/mL)	INS (mU/L)
Blank group	10	—	19.68 ± 1.19	26.18 ± 1.57	27.39 ± 2.66
Model group	10	—	17.59 ± 1.19$^{\Delta\Delta}$	29.97 ± 1.83$^{\Delta\Delta}$	30.60 ± 0.33$^{\Delta\Delta}$
Dacin-35 group	10	0.339	19.47 ± 1.14**	26.45 ± 1.64**	27.96 ± 0.45**
Dodder total flavone	10	200	19.40 ± 1.18**	26.81 ± 1.26**	28.02 ± 1.18**
	10	100	19.12 ± 1.02**	26.95 ± 1.39**	28.32 ± 1.48**
	10	50	19.02 ± 1.08**	27.33 ± 1.93**	28.85 ± 0.90**

Note: Compared to the blank group, $^{\Delta}$represents $P < 0.05$, $^{\Delta\Delta}$represents $P < 0.01$; Compared to the model group, *represents $P < 0.05$, **represents $P < 0.01$.

Table 4
Effect of dodder total flavone on TNF-α and IGF-l protein expression in ovariary of PCOS rat models (mean light density).

Group	n	Dose (mg/kg)	TNF-α	IGF-1
Blank group	10	–	0.260 ± 0.018	0.203 ± 0.008
Model group	10	–	0.307 ± 0.015$^{\Delta\Delta}$	0.222 ± 0.009$^{\Delta\Delta}$
Dacin-35 group	10	0.339	0.256 ± 0.010**	0.201 ± 0.006**
Dodder total flavone	10	200	0.253 ± 0.015**	0.201 ± 0.005**
	10	100	0.268 ± 0.023**	0.206 ± 0.010**
	10	50	0.258 ± 0.019**	0.210 ± 0.011*

Note: Compared to the blank group, $^{\Delta}$represents $P < 0.05$, $^{\Delta\Delta}$represents $P < 0.01$; Compared to the model group, *represents $P < 0.05$, **represents $P < 0.01$.

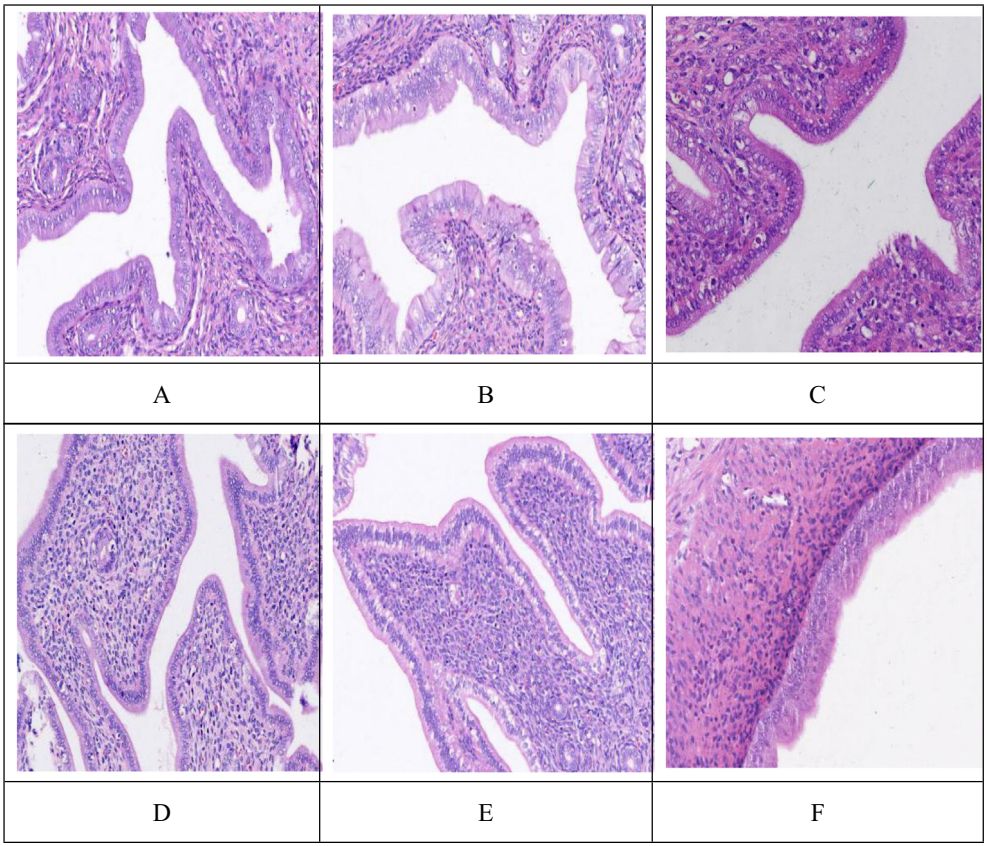

Fig. 1. Effect of dodder total flavone on uterine tissue of PCOS rat models induced by DHEA combined HCG (A: Blank group; B: Model group; C: Dacin-35 group; D: Large dose dodder total flavone group; E: Middle dose dodder total flavone group; F: Small dose dodder total flavone group).

Table 5
Effects on uterine tissue of PCOS rat models.

Group	n	Dose (mg/kg)	–	+	++	+++
Blank group	10	–	10	0	0	0
Model group	10	–	0	1	7	2
Dacin-35 group	10	0.339	8	1	1	0
Dodder total flavone	10	200	6	3	1	0
	10	100	5	3	2	0
	10	50	4	4	1	1

"–" normal, endometrial gland epithelial cells in a single column, the surface of the villi arranged neatly; "+" glandular epithelial cells are basically normal, occasional inflammatory cells, slight chronic inflammation; "++" glandular epithelial cells were mildly proliferated, a small number of uterine cavity or deep gland were dilated, most inflammatory cells were observed, and moderate chronic inflammation was observed; "+++" glandular epithelial cell hyperplasia, most uterine cavity or deep gland dilate, a large number of inflammatory cells, moderate-severe chronic inflammation.

6. Discuss

The clinical manifestations of PCOS are highly heterogeneous, and it is a chronic endocrine disorder with multiple causes and systems. PCOS can cause non-ovulation infertility and ovulation disorder infertility, cause long-term complication such as amenorrhea, glucose and lipid metabolism disorder, endometrial hyperplasia or cancer, endanger the long-term health of the body (Gui et al., 2018; Siddamalla et al., 2018). TCM emphasizes the combination of dialectic and disease differentiation. There is no unanimous con-

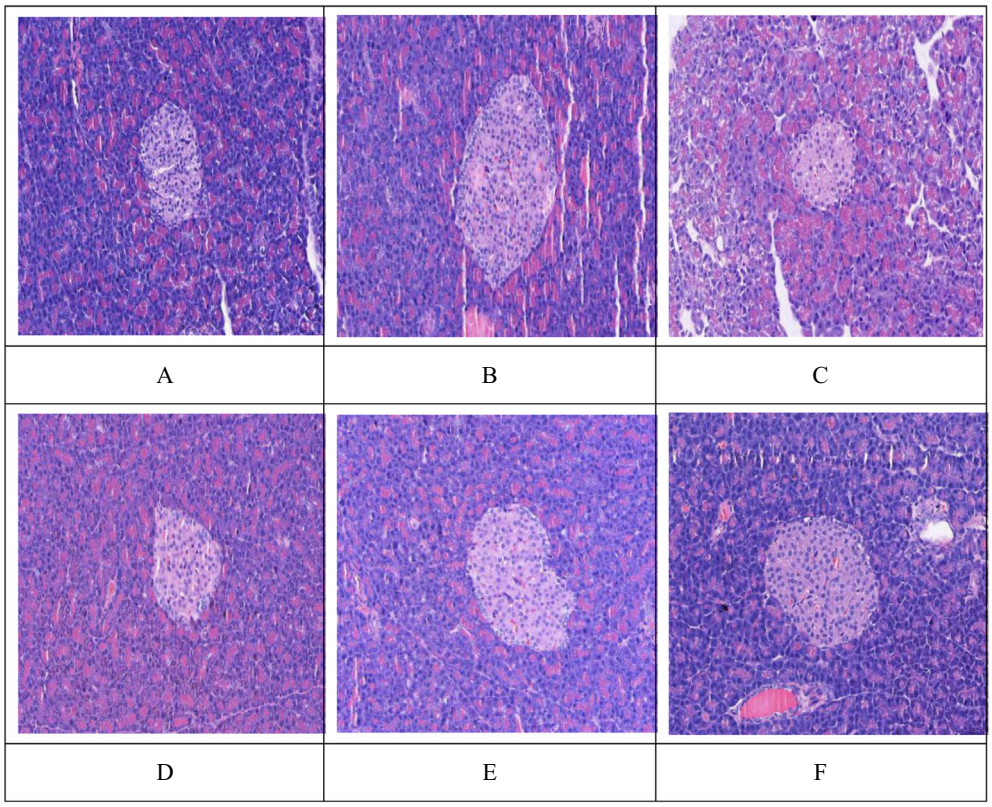

Fig. 2. Effect of dodder total flavone on islet tissue of PCOS rat models induced by DHEA combined HCG (A: Blank group; B: Model group; C: Dacin-35 group; D: Large dose dodder total flavone group; E: Middle dose dodder total flavone group; F: Small dose dodder total flavone group).

Table 6
Effect of dodder total flavone on islet tissue of PCOS rat models induced by DHEA combined HCG ($\bar{x} \pm s$).

Group	n	Dose (mg/kg)	Islet diameter (μm)
Blank group	10	–	142.28 ± 12.78
Model group	10	–	241.52 ± 18.32$^{\Delta\Delta}$
Dacin-35 group	10	0.339	153.07 ± 8.80**
Dodder total flavone	10	200	177.24 ± 8.15**
	10	100	178.83 ± 9.72**
	10	50	182.08 ± 15.22**

Note: Compared to the blank group, $^{\Delta}$represents $P < 0.05$, $^{\Delta\Delta}$represents $P < 0.01$; Compared to the model group, *represents $P < 0.05$, **represents $P < 0.01$.

clusion on the etiology and pathogenesis of PCOS. Most scholars believe that renal, liver and spleen functions jointly influence PCOS, and mostly treated from the perspective of kidney (Wang et al., 2017). Common TCM syndromes of PCOS include kidney deficiency and blood stasis, kidney deficiency and sputum stasis, kidney deficiency and phlegm dampness, and phlegm dampness block, kidney deficiency can be seen almost throughout the PCOS. PCOS belongs to the categories of "infertility", "amenorrhea", "avulsion" and "menstrual disorder" in TCM. Systematic cluster analysis showed that gynecological diseases such as female infertility, menstrual disorder, amenorrhea, ovulation free, polycystic ovary syndrome in Chinese and western medicine were the first category of diseases related to kidney deficiency (Liu et al., 2012), which further verified the essence of kidney deficiency in PCOS patients. Throughout the history of Chinese medicine for various gynecological diseases, there is no lack of application of Chinese medicine for tonifying kidney. In the treatment of PCOS, TCM mostly use the methods of tonifying kidney, which reflects the principle of "regulating menstruation, must tonify kidney".

The classification of PCOS in Chinese medicine and the rule of drug use were summarized in the literature, the results showed that kidney deficiency was widely considered to be the main pathogenic factor of the disease. And kidney-tonifying Yang medicinefrequently appeared in the treatment of the disease. However, the clinical application of tonifying kidney-yang medicine in PCOS is often combined with other drugs in the form of prescription. In order to further clarify the rationality of kidney-tonifying Chinese medicine intervention and treatment of PCOS, and confirm its efficacy, it is necessary to study single Chinese medicine. In view of this phenomenon, it is proposed to use kidney-tonifying Yang medicine to treat gynecological diseases. Based on different animal models to confirm the curative effect of kidney-tonifying Yang medicine commonly used in clinical prescription, it has been found that Chinese medicine for kidney-yang has significant effects on perimenopausal syndrome (Fu et al., 2018), perimenopausal depression (Liu et al., 2012), uterine dysplasia and dysmenorrhea. Dodder is a kind of traditional Chinese medicine commonly used for kidney-tonifying, which has many pharmacological effects, such as regulating immunity, anti-aging, protecting liver, regulating reproductive system, protecting cardiovascular and cerebrovascular, lowering blood glucose. Flavonoids are its main active components (Miao et al., 2017). In this study, the polycystic ovary syndrome model of rats was established by DHEA combined with HCG to explore the intervention effect of total flavonoids of dodder on PCOS rats.

PCOS patients have certain follicular excretion disorder, which to some extent leads to ovarian hyperplasia (Zhang et al., 2016; Wang et al., 2012) and increased uterine volume. The total flavonoids of dodder could reduce ovarian and uterine indices, so as to improve the PCOS rat model. High LH/FSH (follicular stimulating hormone) level is one of the typical characteristics of PCOS. High LH (luteinizing hormone) inhibits follicular stimulating hormone

(FSH) function, making granulocytes flavinized, small sinus follicle development stopped, and ovarian polycystic changes. The increase of serum PRL and LH/FSH ratio is a manifestation of endocrine disorder, and the reduction of serum PRL and LH/FSH ratio can regulate the endocrine disorder in PCOS patients, thereby delaying the progress of the disease and improving the disease (Yin et al., 2018). Because PCOS ovaries do not ovulate regularly, they cannot secrete progesterone, and the amount of progesterone is lower than that of normal patients (Emaduldin and Enas, 2018; Sun et al., 2018; Brosens and Benagiano, 2015). Moreover, PCOS patients are often accompanied by different degrees of insulin resistance (Xu and Xiong, 2016; Wei and Burren, 2017), and their serum INS level are increased to different degrees. PCOS is a low-grade inflammatory reaction, which can damage the capillary endothelium and pancreatic islet cells, and damage the ovary, leading to polycystic changes and abnormal hormone secretion. Therefore, improving the inflammatory response is the key to the treatment of PCOS (Jiang et al., 2018). Inflammatory factors in PCOS patients can indirectly promote androgen synthesis by inducing insulin resistance (IR), among which TNF-α is one of inflammatory cytokines studied most (Lee et al., 2018). High level insulin-like growth factor-1 (IGF-1) can enhance the activity of cytochrome P450c17 enzyme, stimulate the follicular membrane to synthesize excessive androgens, and aggravate hyperandrogenemia (Chen et al., 2017; Zhong et al., 2018). Pregnancy rate is related to endometrial receptivity and endometrial thickness, and endometrial dysplasia leads to a reduction in pregnancy rate (Zeng et al., 2018; Azemi et al., 2018; Lungu et al., 2017; Kumar and Kumar, 2017; Gokharman et al., 2017; Gao et al., 2017; Xu et al., 2018). The results showed that total flavonoids of dodder could reduce serum LH/FSH ratio, PRL and INS levels and increase serum P levels, decrease the levels of TNF-α and IGF-1 proteins in ovarian tissue. Which indicating that it can achieve the intervention effect on PCOS model rats by improving the endocrine, inflammatory reaction, insulin resistance, uterus and islet pathological changes.

In conclusion, total flavonoids of dodder can decrease the ovary and uterus index, affect the secretion of serum hormones (LH/FSH, PRL, INS and P), regulate ovarian protein expression (TNF-α and IGF-1); improving endometrial hyperplasia, inflammatory infiltration and islet tissue lesions, so as to achieve the protective effect on PCOS rat models induced by DHEA combined HCG.

Acknowledgement

This research was financially supported by NSFC (Natural Science Foundation of China: 81274154) and Zhong Yuan Scholars (162101510003).

References

Azemi, A.K., Abd Rahim, M.H., Mamat, S.S., Jais, A.M.M., Zakaria, Z.A., 2018. Pakistan J. Pharm. Sci. 31 (1), 143–151.
Brosens, I., Benagiano, G., 2015. Menstrual preconditioning for the prevention of major obstetrical syndromes in polycystic ovarysyndrome. Am. J. Obstetr. Gynecol. 213 (4), 488–493.
Chen, Y., Liang, H., Wang, N., 2017. The effect of Bushen Tiaogan decoction on serum TGF-β1 IGF-1, TNF-a and hormone levels in rats with polycystic ovary syndrome. Shaanxi J. Trad. Chin. Med. 38 (08), 1148–1150.
Daghestani, M.H., 2018. Evaluation of biochemical, endocrine, and metabolic biomarkers for the early diagnosis of polycystic ovary syndrome among non-obese Saudi women. Int. J. Gynecol. Obstetr. 142 (2), 162–169.
Elbohoty Ahmed, E.H., Mohamed, Amer, Abdelmoaz, M., 2016. Clomiphene citrate before and after withdrawal bleeding for induction of ovulation in women with polycystic ovary syndrome: Randomized cross-over trial. Obstetr. Gynaecol. Res. 42 (8), 966–971.
Fu, Z.N., Wei, Z.Z., Miao, M.S., 2018. Effects of total flavonoids of raspberry on perimenopausal model in mice. Saudi J. Biol. Sci. 25 (3), 487–492.
Gao, W., Wang, Y., Wang, W., Shi, L., 2017. Saudi Pharm. J. 25 (4), 548–555.
Giandalia, A, Pappalardo, M.A., Russo Giuseppina, T., Romeo, E.L., Alibrandi, A., Di Bari, F., Vita, R., Cucinotta, D., Benvenga, S. Influence of peroxisome proliferator-activated receptor-gamma exon 2 and exon 6 and insulin receptor substrate (IRS)-1 Gly972Arg polymorphisms on insulin resistance and beta-cell function in southern mediterranean women with polycystic ovary syndrome. J. Clin. Transl. Endocrinol., (13) (2018) 1–8.
Gokharman, F.D., Aydin, S., Fatihoglu, E., Kosar, P.N., 2017. Turkish J. Med. Sci. 47 (6), 1770–1777.
Gui, H., Huang, Y., Zhao, Q.Y., 2018. Clinical effect analysis of integrated Chinese and Western medicine for polycystic ovary syndrome. J. New Chinese Med. 50 (06), 142–144.
Huang, Y.M., Kang, K.B., Pan, W., Yuan, R.Z., 2018. Law of drug using in syndrome differentiation of chinese medicine for polycystic ovary syndrome based on data mining. J. New Chinese Med. 50 (06), 60–64.
Huang, C.S., Tan, T.T., Xing, P.T., 2016. Clinical study on the effects of dodder weed on glycolipid metabolism in gestational diabetes mellitus. Modern J. Integr. Trad. Chinese Western Med. 25 (20), 2199–2201.
Jamilian, M., Samimi, M., Mirhosseini, N., Ebrahimi, FA., Aghadavod, E., Talaee, R., Jafarnejad, S., Dizaji, SH., Asemi, Z. The influences of vitamin D and omega-3 co-supplementation on clinical, metabolic and genetic parameters in women with polycystic ovary syndrome. J. Affect. Disord. (238) (2018) 32–38.
Jiang, X.L., Zhang, B., HaiY, Hou, Teng, F., Xiande, Ma., Meijun, Lyu, Ying, Wang, Chunli, Ch.u., Xiaoqian, Dong, Lide, Zhang, 2018. Influence of metformin combined with Gexia Zhuyu Tang on inflammatory factors and sex hormone level in rats with polycystic ovarian syndrome. J. Beijing Univ. Trad. Chin. Med. 41 (01), 65–70.
Kumar, D., Kumar, S., 2017. Indian J. Pharm. Sci. 79 (6), 1001–1007.
Lee, H., Oh, J.Y., Sung, Y.A., 2018. Adipokines, insulin-like growth factor binding protein-3 levels, and insulin sensitivity in women with polycystic ovary syndrome. Korean J. Intern. Med. 23 (4), 456–463.
Leng, Q., Wei, Z.L., 2018. Research progress in the etiology, pathogenesis and treatment of polycystic ovary syndrome. J. Int. Reprod. Health/Family Plann. 37 (01). 57–61+75.
Li, Y., Li, R.Q., Ou, S.B., Zhang, N.F., Chen, X.H., Zhang, Q.X., Yang, D.Z., 2018. Comparisons of secretive profile and diagnostic power of anti-mullerian hormone in women with polycystic ovaries with and without hyperandrogenism. J. Pract. Obstetr. Gynecol. 2014 30 (2), 111–115.
Liu, S.C., Liu, Y., Song, H., Zheng, H.X., 2012. Study on nephrotic deficiency syndrome syndrome spectrum based on literature co-word analysis. World Sci. Technol. (Modernization of Traditional Chinese Medicine and Materia Medica) 14 (02), 1408–1412.
Liu, H.X., Zhao, Y.N., Huo, J.N., 2018. Effects of dehydroepiandrosterone and high-fat diet on induction of a rat model of polycystic ovary syndrome. J. China Med. Univ. 47. 961–963+974..
Lungu, M., Romila, A., Nechita, A., Tutunaru, D., Marian, C.B., 2017. ACTA Medica Mediterranea 33 (3), 385–392.
Miao, M.S., Tian, S., Guo, L., Bai, M., Fang, X.Y., Liu, S.Y., 2017. The effect of curculigoside on mouse model of perimenopausal depression. Saudi J. Biol. Sci. 24 (8), 1894–1902.
Miao, M.S., Peng, M.F., Yan, X.L., 2018. Effects of dodder total flavone on polycystic ovary syndrome rat models. Chin. J. Exp. Trad. Med. Formulae, 1–8. https://doi.org/10.13422/j.cnki.syfjx.20190506.
Ren, X.Q., Zheng, G.Z., Su, H., Zhang, X., Zhai, W.S., Tang, J.F., Li, W.X., Ding, Y., 2018. Influence of flavonoids from Cuscutae Semen on cell cycle arrest, apoptosis and protein expression of spermatogenic cells induced by multi-glycoside from Tripterygium Wilfordii. Drug Eval. Res. 41 (01), 55–60.
Rezvan, N., Moini, A., Gorgani-Firuzjaee, S., Hosseinzadeh-Attar, M.J., 2018. Oral quercetin supplementation enhances adiponectin receptor transcript expression in polycystic ovary syndrome patients: a randomized placebo-controlled double-blind clinical trial. Cell J. 19 (4), 627–633.
Seyam, Emaduldin, Hefzy, Enas, 2018. Long-term effects of combined simvastatin and metformin treatment on the clinical abnormalities and ovulation dysfunction in single young women with polycystic ovary syndrome. Gynecol. Endocrinol.: Off. J. Int. Soc. Gynecol. Endocrinol., 1–8
Siddamalla, S., Reddy, T.V., Govatati, S., Guruvaiah, P., Deenadayal, M., Shivaji, S., Bhanoori, M., 2018. Influence of tumour suppressor gene (TP53, BRCA1 and BRCA2) polymorphisms on polycystic ovarysyndrome in South Indian women. Eur. J. Obstetri. Gynecol. Reprod. Biol. 227, 13–18.
Sun, P.P., Wang, G.H., Gao, D., 2018. Clinical effects of metformin combined with clomiphene on infertility of patients with polycystic ovary syndrome and its influence on endocrine. Anhui Med. Pharm. J. 22 (03), 518–522.
Wang, Q., Kim, J.Y., Xue, K., Liu, J.Y., Leader, A., Tsang, B.K., 2012. Chemerin, a novel regulator of follicular steroidogenesis and its potential involvement in polycystic ovarian syndrome. Endocrinology 153 (11), 5600–5611.
Wang, M.J., Yu, J., Huang, N., Zhou, Y.N., Wu, M., 2017. Research on features of TCM syndrome elements in polycystic ovary syndrome. J. Yunnan Univ. Trad. Chin. Med. 40 (05). 27–29+48.
Wei, C., Burren, C.P., 2017. Diagnostic and management challenges from childhood, puberty through to transition in severe insulin resistance due to insulin receptor mutations. Pediatr. Diabetes 18 (8), 835–838.
Xu, K., He, G., Qin, J., Cheng, X., He, H., Zhang, D., Peng, W., 2018. Saudi J. Biol. Sci. 25 (4), 811–815.
Xu, L., Xiong, Y., 2016. Effects of ethinylestradiol and cyproterone acetate combined with metformin on ovulation induction of patients with polycystic ovarian syndrome. China Pharm. 27 (24), 3368–3370.

Yin, Y., Zhang, Y.C., Zhang, H., Jiang, D.S.G., Guo, G.R., 2018. Clinical therapeutic effects of acupuncture combined with Chinese herbal medicine on infertility of polycystic ovary syndrome in the patients with ovulation induction with letrozole. Chin. Acupuncture Moxibustion 38 (01), 27–32.

Zeng, Q., Wang, Y.X., Zhang, H.X., 2018. Invigorating kidney and resolving phlegm method of intervention on kidney yang deficiency PCOS ovulation disorder infertility patients with endometrial receptivity and ovulation. Shaanxi J. Trad. Chin. Med. 39 (03), 334–337.

Zhang, Y.L., Pan, W., Wang, Y.J., 2016. Advances in Chinese and western medicine on pathogenesis of polycystic ovary syndrome. Chinese J. Basic Med. Trad. Chin. Med. 22 (07), 1004–1006.

Zhao, S.X., Liu, H.L., Jiang, H., 2018. Study on the mechanism of Wnt/beta-catenin signaling pathway to investigate the bone metabolism of rats in ovarian rats. J. Clin. Exp. Med. 17 (01), 25–28.

Zhong, Z.H., Li, F., Li, Y.Y., Qin, S., Wen, C.L., Fu, Y.Y., Xiao, Q., 2018. Inhibition of microRNA-19b promotes ovarian granulosa cell proliferation by targeting IGF-1 in polycystic ovary syndrome. Mol. Med. Rep. 17 (4), 4889–4898.

Zuo, L., Fu, Y.J., 2018. Etiology and treatment progress of polycystic ovary syndrome. Chongqing Med. 47 (09), 1247–1250.