doi: 10.13241/j.cnki.pmb.2015.12.002

离体子宫张力测定在药物生殖毒性研究中的应用 *

崇立明 姜 娟 杨 阳 马爱翠 苏 欣 周 莉 孙祖越△

(上海市计划生育科学研究所 中国生育调节药物毒理学检测中心 上海 200032)

摘要 目的:建立药物生殖毒性研究中孕鼠离体子宫平滑肌张力的测定方法。**方法:**SD 大鼠于妊娠第 6~15 天(GD6-15)给予受试物,在妊娠第 20 天(GD20)取子宫平滑肌,分别给予不同浓度缩宫素和硫酸镁溶液刺激,选择试验的最佳条件。在此基础上,测定不同剂量组孕鼠离体子宫平滑肌张力的变化情况,采用 RM-6240BD 多道生理信号采集处理系统分析数据,统计结果。**结果:**选择 0.007 U/mL 缩宫素、0.008 mol/mL 硫酸镁为最佳刺激浓度。随着受试物剂量的增加,加入缩宫素后,各组妊娠子宫平滑肌的频率和张力均呈逐渐上升的趋势,而加入硫酸镁后,各剂量组妊娠子宫平滑肌的活动幅度、频率和张力均呈现降低的趋势,但各剂量组与溶媒对照组相比均未见明显差异(P>0.05)。**结论:**本研究建立了孕鼠离体子宫平滑肌张力的测定方法,该方法可以更好的反映受试物对孕鼠子宫肌的毒性作用,更加全面的评价受试物的生殖毒性。

关键词:生殖毒性;孕鼠;离体;子宫平滑肌;张力

中图分类号:Q95-3;R96 **文献标识码:**B **文章编号:**1673-6373(2015)12-2205-04

Application of Isolated Uterine Tension in the Study of Reproductive Toxicity of Drugs*

CHONG Li-ming, JIANG Juan, YANG Yang, MA Ai-cui, SU Xin, ZHOU Li, SUN Zu-yue△

(Shanghai Institute of Planned Parenthood Research &National Evaluation Centre for Toxicology of Fertility Regulating Drugs, Shanghai, 200032, China)

ABSTRACT Objective: To establish a method of detecting the tension of isolated pregnant rat myometrium strips in the reproductive toxicity of drugs. **Methods:** SD rats were treated with drugs on the 6th-15th(GD6-15) of gestation, the uterine smooth muscle was taken on the 20th(GD20) of gestation and treated with different concentrations of oxytocin and magnesium sulfate solution to choose the best conditions for testing. Then the effect of different dosages of drugs on the activity of isolated uterus smooth muscle of pregnant rats were observed by RM-6240BD biological signaling system. **Results:** 0.007 U/mL magnesium sulfate and 0.008 mol/mL oxytocin were selected as the best concentration. With the increase of drug dosage, the frequency and tension of pregnant uterine smooth muscle of each group all showed a rising trend. While after the treatment with magnesium sulfate, the amplitude, frequency and tension of uterine smooth muscle of each dose group tends to decrease, but no significant difference was found between each dose group and control group (P>0.05). **Conclusion:** A method for the determination of muscle tension from isolated uterus of pregnant rats was established, which could better reflect the toxicity of the test substances on pregnant rats' uterine muscle and comprehensively evaluate the reproductive toxicity.

Key words: Reproductive toxicity; Pregnantrats; Isolated tissue; Uterine smooth muscle; Tension

Chinese Library Classification (CLC): Q95-3; R96 Document code: B

Article ID: 1673-6373(2015)12-2205-04

前言

离体子宫平滑肌是生物活性组织,通过人工模拟的生理环境测定药物对子宫平滑肌舒张和收缩变化的影响,同时采用计算机生物信号采集处理系统,使子宫平滑肌收缩的复杂信号量化,绘制出给药前后子宫平滑肌收缩的图形并测得数据,为实验提供更加客观和准确定量的分析方法。

无论对于妊娠动物还是非妊娠动物而言,经典的离体子宫药理学研究通常的方法是取子宫平滑肌片段,在体外加入不同浓度的药物,以测定给药前后子宫平滑肌张力的变化,可以测定药物对受体的亲和力,决定药物是激动剂还是拮抗剂,并能分析药物与受体的构效关系, 反映药物在器官水平上的作用。但在胚胎 - 胎仔发育毒性试验中,采用体外给药的方法无法模拟体内实验中受试物对妊娠子宫的影响,因此我们建立了动物体内给药模型,并在体外检测子宫平滑肌张力的方法,该方法未见文献报道。本研究根据《药物生殖毒性研究技术指导原则》

* 基金项目:国家十二五 " 重大新药创制 " 科技重大专项(2011ZX09301-005);
上海市实验动物创新行动计划项目(11140901300;11140901303)
作者简介:崇立明(1988-),男,本科,研究实习员,主要研究方向:生殖药理毒理学,电话:18217587105,E-mail:chonglm@163.com
△通讯作者:孙祖越,E-mail:sunzy64@163.com
(收稿日期:2014-10-25 接受日期:2014-11-20)

[1],动物于 GD6-15 接受不同剂量的受试物,利用体内给药妊娠的动物模型,GD20 选取妊娠动物的子宫平滑肌,检测其对刺激的舒张和收缩变化情况，以便同步分析药物对母体的毒性作用。

1 材料与方法

1.1 动物

健康 SD 大鼠,SPF 级，♀ 的体重 180~200 g，♂ 的体重 200~220 g,购自上海西普尔 - 必凯实验动物有限公司,生产许可证 SCXK(沪)2008-0016,使用许可证 SYXK(沪)2008-0027,饲养于上海市计划生育科学研究所(中国生育调节药物毒理检测中心)SPF 级动物房,保持室温 20~26℃ ,相对湿度 40~70%,自由进食饮水。

1.2 药品与试剂

缩宫素注射液(上海禾丰制药有限公司,10 U/mL,实验前稀释至 2 U/mL);硫酸镁溶液(配制 0.96 mol/L);De-Jalon's 液配方:NaCl 9 g,KCl 0.42 g,$NaHCO_3$ 0.5 g,$CaCl_2$ 0.06 g，葡萄糖 0.5 g,蒸馏水 1000 mL[2]。

1.3 仪器

RM-6240BD 多道生理信号采集处理系统;JZJ01H 型肌肉张力换能器;离体肠管及热板恒温器(均购自成都仪器厂)。

1.4 模型制备

按雌雄 2:1 于当日下午 16:00 合笼交配,次日上午 8:00 以后阴道涂片检查,查到精子或阴栓确定为妊娠第 0 天,随机分组,妊娠第 6~15 天给予不同浓度受试物。在妊娠第 20 天,采用 3%戊巴比妥钠麻醉孕鼠,剖腹取子宫,快速清除胎仔和胎盘组织,取其子宫体部平滑肌,修剪成约 3× 10mm 大小[3]。

1.5 子宫平滑肌张力测定

开启多道生理信号采集处理系统,调零,调节水浴温度为 35℃ ,连续通入混合氧(1～2 个气泡 / 秒);取上述方法制备的子宫平滑肌条,一端穿线固定于离体器官恒温水浴装置的浴槽底部,另一端穿线与记录装置相连;将已连接的子宫肌浸没于浴槽,浴槽中盛有 6 mL 新鲜配置的 De-Jalon's 液,给予子宫肌 1 g 的负荷,并保持;静置 15 min 待波形稳定后,记录约 5 min 正常波形;在浴槽中加入 21 μL 缩宫素注射液,记录约 5 min 波形;清洗浴槽 2-3 次后,加 6 mL 营养液稳定 2 min,加入 50 μL 硫酸镁溶液,记录约 5 min 波形;保存文件,分析波形。

1.6 统计学处理

以 RM-6240BD 生物机能实验系统描记子宫平滑肌的收缩活动,测定子宫平滑肌的收缩幅度、频率及张力(幅度× 频率)。实验数据均以均数± 标准差($\bar{x}$± s)表示,采用 SPSS 11.5 统计分析软件处理实验数据，多组间比较采用单因素方差分析,以 P<0.05 为差异有统计学意义。

2 结果

2.1 最佳缩宫素和硫酸镁溶液刺激剂量的确定

给予未接受受试物的孕鼠子宫肌 0~0.01 U/mL 缩宫素时,表现为随剂量升高，离体子宫的收缩增强，当剂量超过 0.01 U/mL 时,随剂量升高,张力持续增高,最后可致强直性收缩。综合选择缩宫素浓度为 0.007 U/mL,可使子宫平滑肌呈较好的节律性收缩。

给予 0~0.01 mol/mL 硫酸镁时,表现为随剂量升高,对离体子宫的抑制作用增强,当剂量超过 0.01 mol/mL 时,离体子宫的收缩明显受到抑制,表现为图形不再波动。综合选择硫酸镁浓度为 0.008 mol/mL,可较好地抑制子宫平滑肌(图 1-3)。

2.2 缩宫素和硫酸镁对受试物处理的妊娠子宫平滑肌幅度、频率和张力的影响

如表 1 所示,未加入任何刺激前,随着受试物剂量的增加,妊娠子宫平滑肌的幅度、频率和张力呈逐渐上升的趋势,但各剂量组与溶媒对照组相比均未见明显差异(P>0.05);加入缩宫素后，各剂量组妊娠子宫平滑肌活动频率和张力有增加的趋势,但各组的幅度、频率和张力与溶媒对照组相比均未见统计学差(P>0.05);加入硫酸镁后,各剂量组妊娠子宫平滑肌的活动幅度、频率和张力均呈现降低的趋势,但与溶媒对照组相比也未见统计学差异(P>0.05)。图 4-6 为给药组孕鼠的部分典型图形。

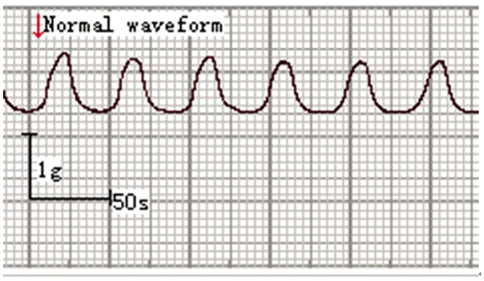

图 1 给予刺激前,孕鼠离体子宫平滑肌波形

Fig.1 Before the spur, the waveform of pregnant rat uterus smooth muscle in vitro

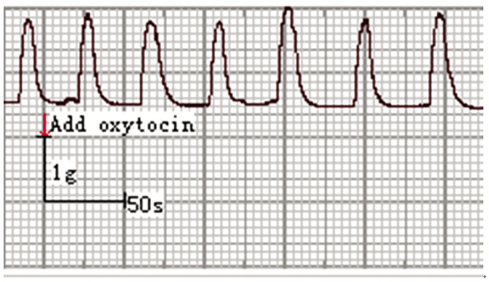

图 2 加入缩宫素,孕鼠离体子宫平滑肌波形

Fig.2 After the treatment with oxytocin, the waveform of pregnant rat uterus smooth muscle in vitro

3 讨论

离体子宫是药理学实验中常用的标本,利用妊娠或非妊娠大鼠子宫平滑肌研究各种药物的影响,已经有不少报道[4-13],主要是离体情况下给予不同浓度的药物,测定离体子宫对不同浓度药物的反应。这种子宫平滑肌的异常收缩或舒张,通常是在药物筛选阶段进行,考察药物的药效作用或药理作用。药物的非临床生殖毒性安全性评价研究所采用的是体内给药动物模型,妊娠期给予不同剂量的药物后,可能会对妊娠子宫的收缩或舒张造成不同程度的影响,从而影响妊娠的结局,研究给予

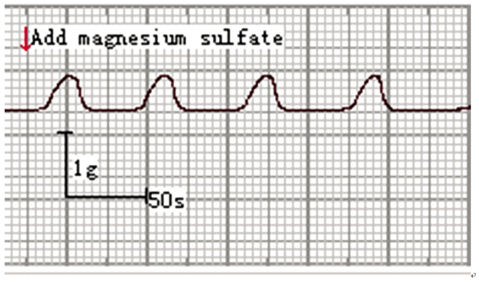

图 3 加入硫酸镁，孕鼠离体子宫平滑肌波形

Fig.3 After the treatment with magnesium sulfate, the waveform of pregnant rat uterus smooth muscle in vitro

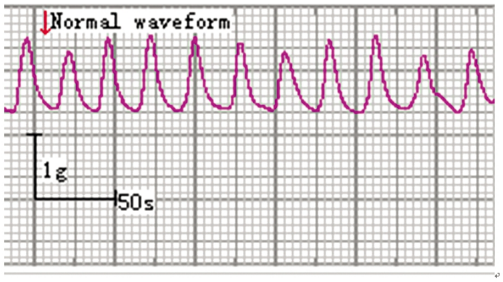

图 4 体内给药后，高剂量组孕鼠离体子宫平滑肌波形，与溶媒对照组相比，幅度、频率和张力有增加的趋势

Fig.4 After the treatment, the waveform of high dose group of pregnant rat uterus smooth muscle in vitro, compared with solvent control group, amplitude, frequency and tension has a tendency to increase

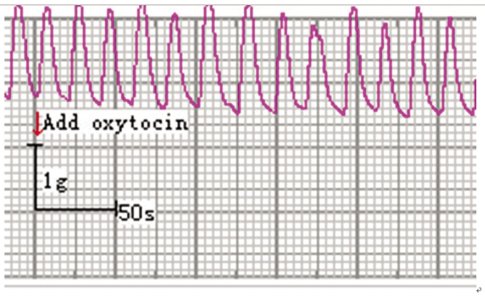

图 5 体内给药后，高剂量组孕鼠离体子宫平滑肌加入缩宫素，与溶媒对照组相比，幅度、频率和张力有增加的趋势

Fig.5 After the treatment, high dose group of pregnant rats in vitro smooth muscle of the uterus to join the oxytocin, compared with solvent control group, amplitude, frequency and tension has a tendency to increase

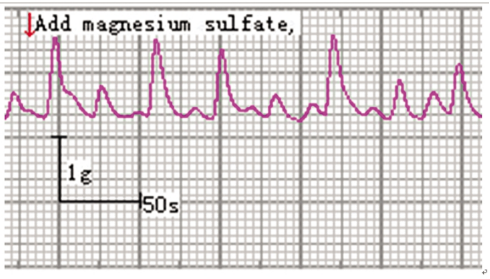

图 6 体内给药后，高剂量组孕鼠离体子宫平滑肌加入硫酸镁，与溶媒对照组相比，幅度、频率和张力有增加的趋势

Fig.6 After the treatment, high dose group of pregnant rats in vitro smooth muscle of the uterus to join magnesium sulfate, compared with solvent control group, amplitude, frequency and tension has a tendency to increase

表 1 缩宫素和硫酸镁刺激对各剂量组子宫平滑肌收缩幅度和频率的影响($\bar{x}\pm s$)

Table 1 Effect of oxytocin and magnesium sulfate on the amplitude and frequency of uterine smooth muscle in different dose group

Groups	Case	Before the spur		Oxytocin stimulates (0.007U/mL)		Magnesium sulfate stimulate (0.008 mol/mL)	
		Amplitude (g)	Frequency (次 /200S)	Amplitude (g)	Frequency (次 /200S)	Amplitude (g)	Frequency (次 /200S)
Solvent control group	14	0.630± 0.122	3.6± 1.9	0.649± 0.082	4.9± 2.3	0.557± 0.086	2.1± 1.8
Low dose group	13	0.622± 0.119	3.8± 1.7	0.695± 0.140	5.5± 2.0	0.532± 0.091	2.8± 1.9
Middle dose group	12	0.657± 0.144	4.5± 1.3	0.670± 0.132	5.8± 1.7	0.552± 0.138	3.1± 1.0
High dose group	15	0.664± 0.151	4.9± 2.1	0.695± 0.158	6.3± 2.6	0.570± 0.116	3.6± 1.7

药物后的妊娠子宫收缩就显得非常重要。因此，根据药物的特点，我们在评价生殖毒性时增加了子宫平滑肌的检测[14,15]。但在生殖毒性研究中，评价这种子宫平滑肌的异常收缩或舒张可能造成不良的后果，几乎未见报道。本研究在药理学研究的基础上，分别探讨了加入缩宫素和硫酸镁后，不同剂量组妊娠动物模型离体子宫平滑肌张力的变化。

缩宫素[16,17]的结构与人体垂体后叶产生的天然催产素相同，能直接兴奋子宫平滑肌，刺激其节律性收缩，为子宫平滑肌兴奋药。本研究利用子宫平滑肌的特点，采用缩宫素注射液，刺激离体子宫收缩。经过预试验，选择 0.007 U/mL 作为统一的试验标准剂量，可使妊娠离体子宫平滑肌呈较好地节律性收缩。硫酸镁[18,19]中的镁离子能直接作用于子宫肌细胞，可拮抗钙离子对子宫肌收缩作用，抑制子宫收缩，为子宫平滑肌抑制药。在本实验室条件下，硫酸镁终浓度为 0.008 mol/mL 时，能较好地抑制妊娠离体子宫平滑肌的收缩。

本研究方法为配合胚胎 - 胎仔发育毒性试验所进行的子宫平滑肌张力检测，该实验中受试物对母鼠的毒性主要表现为中高剂量组母鼠体重增长缓慢和高剂量导致的宫外增重减少，对胚胎 - 胎仔发育毒性表现为高剂量活胎率降低，吸收胎率增加。实验结果表明子宫平滑肌对缩宫素和硫酸镁的刺激显示出剂量 - 反应增加趋势，考虑到受试物中含有红花，红花有活血化瘀、使子宫紧张性和兴奋性增加的药理作用[20]。故可认为增加该指标的检测可以更好的反映受试物的生殖毒性作用，更加全面的评价受试物的特点。

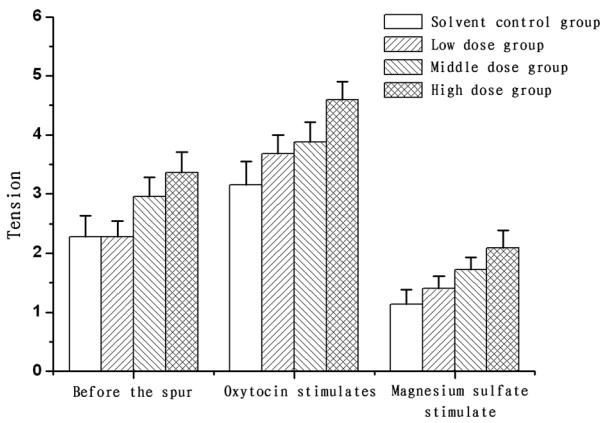

图 7 缩宫素和硫酸镁刺激对子宫平滑肌张力(幅度× 频率)的影响

Fig.7 The effect of oxytocin and magnesium sulfate stimulation on the tension of uterine smooth muscle(Amplitude× Frequency)

参 考 文 献(References)

[1] 国家食品药品监督管理局. 药物生殖毒性研究技术指导原则[M]. 北京:中国医药科技出版社, 2006- 2007: 143-168
State Food and Drug Administration. Guidelines for the testing of drugs reproductive toxicity studies [M]. Beijing: Chinese medical science and technology press, 2006-2007: 143-168

[2] 国家药典委员会. 中国药典[S]. 北京: 中国医药科技出版社, 2010
Chinese pharmacopoeia commission. Chinese Pharmacopoeia [S]. Beijing: Chinese medical science and technology press, 2010

[3] 徐叔云. 药理实验方法学[M] . 北京:人民卫生出版社, 1991: 1467
Xu Shu-yun. Methodology of Pharmacological Experiment [M]. Beijing: People's medical publishing house, 1991: 1467

[4] 韩继超. 槟榔次碱对未孕大鼠离体子宫平滑肌运动的影响[J]. 中华中医药学刊, 2008, 26(2): 379-380
Han Ji-chao. Study on Effect of Arecaidine on the Isolated Uterus of Rats [J]. Chinese archives of traditional chinese medicine, 2008, 26 (2): 379-380

[5] 李伟霞, 华永庆, 唐于平, 等. 归芎药对对小鼠离体子宫收缩活动的影响[J]. 南京中医药大学学报 2010, 26(2): 120-122
Li Wei-xia, Hua Yong-qing, Tang Yu-ping, et al. Effects of Radix Angelicae Sinensis and Rhizoma Chuanxiong on Mouse Uterine Contract ions in Vitro [J]. Journal of Nanjing TCM University, 2010, 26(2): 120-122

[6] 訾慧, 朱会, 张振秋. 吴茱萸当归药对乙醇提取物对大鼠离体子宫平滑肌收缩的影响[J]. 中华中医药学刊, 2009, 27(4): 832-833
Zi Hui, Zhu Hui, Zhang Zhen-qiu. Research on the decreasing function of evodia rutaecarpa angelica sinensis ethanol extract on contractile activity of uterine smooth muscle strips of virginal rats[J]. Chinese archives of traditional chinese medicine, 2009, 27 (4): 832-833

[7] 王淑英, 吴银萍, 王建刚, 等. 西地那非对鼠子宫平滑肌及微循环作用的实验研究[J]. 中国妇幼保健, 2010, 25: 3632-3636
Wang Shu-ying, Wu Yin-ping, Wang Jian-Gang , et al. Experimental study on the effect of sildenafil on rat uterus smooth muscles and micro-circulation[J]. Department of Pharmacology, 2010, 25: 3632-3636

[8] 严作廷, 王东升, 杨国林, 等. 中药产复康对大鼠离体子宫平滑肌收缩功能的影响[J]. 中兽医医药杂志, 2010, 1: 10-13
Yan Zuo-ting, Wang Dong-sheng, Yang Guo-lin, et al. Effects of Chinese herbal medicine Chanfukang on in vitro uterine smooth muscle contraction in rats[J]. JTCVM, 2010, 1: 10-13

[9] 张建英, 楚更五, 刘秀萍, 等. 当归散对正常和缩宫素处理大鼠离体子宫活动力的影响[J]. 中国实验方剂学杂志, 2011, 17(7): 201-203
Zhang Jian-ying, Chu Geng-wu, Liu Xiu-ping, et al. Effects of Danggui San on isolated smooth muscle of uterus and uterine smooth muscle strips stimulated by oxytocin in rats [J]. Chinese Journal of Experimental Traditional Medical Formulae, 2011, 17(7): 201-203

[10] 华永庆,丁爱华,段金廒, 等. 当归芍药散对缩宫素诱导的大鼠在体子宫收缩及血管舒缩因子的影响[J]. 中草药, 2013, 44(4): 459-462
Hua Yong-qing, Ding Ai-hua, Duan Jin-ao, et al. Effects of Danggui Shaoyao Powder on oxytocin induced in vivo uterine contractions and vasomotor factors of rats [J]. Chinese Traditional and Herbal Drugs, 2013, 44(4): 459-462

[11] 李丹, 谢晓芳, 彭成, 等. 益母草注射液提取物对流产大鼠离体子宫的影响[J]. 药物评价研究, 2014, 37(1): 21-24
Li Dan, Xie Xiao-fang, Peng Cheng, et al. Effects of extract from Leonurus japonicus Injection on uterus of rats after abortion [J]. Drug Evaluation Research, 2014, 37(1): 21-24

[12] 仲伟国, 朱海. 罗库溴铵和顺式阿曲库铵对分娩期大鼠离体子宫平滑肌收缩的影响[J]. 第二军医大学学报, 2011, 32(6): 677-679
Zhong Wei-guo, Zhu Hai. Effect of rocuronium and cis-atracuriumon on contraction of isolated uterine smooth muscle in rat of labor Stages [J]. Academic Journal of Second Military Medical University, 2011, 32(6): 677-679

[13] 黄聪, 王建, 王世宇, 等. 合成冰片对非孕大鼠离体子宫平滑肌运动的影响[J]. 中药药理与临床, 2013, 29(3): 104-106
Huang Cong, Wang Jian, Wang Shi-yu, et al. Effects of borneol on the non-pregnant rat uterine smooth muscle in vitro[J]. Pharmacology and Clinics of Chinese Materia Medica, 2013, 29(3): 104-106

[14] 孙祖越, 周莉, 闫晗, 等. 如何成功开展药物非临床生殖毒性试验[J]. 中国新药杂志, 2011, 20(22): 2195-2204
Sun Zu-yue, Zhou-Li, Yan-Han, et al. How to successfully carry out nonclinical reproductive toxicity study on new drugs [J]. Chinese Journal of New Drugs, 2011, 20(22): 2195-2204

[15] 孙祖越, 周莉, 闫晗, 等. 药物非临床生殖发育毒性试验中逐案原则的建议[J]. 中国新药杂志, 2012, 21(8): 836-843
Sun Zu-yue, Zhou-Li, Yan-Han, et al. A proposal for case-by-case principle in reproductive and developmental toxicity studies [J]. Chinese Journal of New Drugs, 2012, 21(8): 836-843

[16] 朱爱民,李昇刚,扆雪涛,等. 缩宫素生物测定试验中离体子宫的稳定性研究[J].中国药事, 2011, 25(10): 988-990
Zhu Ai-min, Li Sheng-gang, Yi Xue-tao, et al. Stability studies of in vitro uterus on biological value test of oxytocin [J]. Chinese Pharmaceutical Affairs, 2011, 25(10): 988-990

[17] 章新华, 王怀良. 大鼠离体子宫平滑肌收缩活性测定方法研究[J]. 中国医科大学学报, 2005, 34(5): 437-438
Zhang Xin-hua, Wang Huai-liang. Study on the determination method of rat uterine smooth muscle contraction in vitro activity [J]. Journal of China Medical University, 2005, 34(5): 437-438
(下转第 2301 页)

2012, 92(25): 1729-1730
Zuo Wen-li. The interpretation of the diagnosis and treatment consensus of cesarean scar pregnancy[J]. Natl Med J China, 2012, 92 (25): 1729-1730

[2] 吴琳琳, 刘欣燕. 剖宫产瘢痕妊娠的临床诊断进展 [J]. 中国计划生育学杂志, 2013, 21(4): 282-285
Wu Lin-lin, Liu Xin-yan. The clinical diagnosis prognosis of caesarean scar pregnancy[J]. Chin J Fam Plann, 2013, 21(4): 282-285

[3] Peng KW, Lei Z, Xiao TH, et al. First trimester caesarean scar ectopic pregnancy evaluation using MRI[J]. Clin Radiol, 2014, 69(2): 123-129

[4] Litwicka K, Greco E. Caesarean scar pregnancy: a review of management options[J]. Curr Opin Obstet Gynecol, 2011, 23(6): 415-421

[5] 邵华江, 马建婷, 徐丽萍, 等. 剖宫产瘢痕妊娠治疗方法与疗效综合分析[J]. 中华医学杂志, 2012, 92(31): 2191-2194
Shao Hua-jiang, Ma Jian-ting, Xu Li-ping, et al. Comprehensive analysis of therapeutic methods and effect on cesarean scar pregnancy [J]. Natl Med J China, 2012, 92(31): 2191-2194

[6] Litwicka K, Greco E. Caesarean scar pregnancy: a review of management options[J]. Curr Opin Obstet Gynecol, 2013, 25(6): 456-461

[7] Timor-Tritsch IE, Monteagudo A. Unforeseen consequences of the increasing rate of cesarean deliveries: early placenta accreta and cesarean scar pregnancy. A review [J]. Am J Obstet Gynecol, 2012, 207(1): 14-29

[8] 欧阳振波, 苏欢欢, 张秋实, 等. 复发性剖宫产术后子宫瘢痕妊娠一例报告及文献复习[J]. 中华妇产科杂志, 2014, 49(4): 298-299
OuYang Zhen-bo, Su Huan-huan, Zhang Qiu-shi, et al. A case of recurrent cesarean scar pregnancy and literature review [J]. Chin J Obstet Gynecol, 2014, 49(4): 298-299

[9] 中华医学会计划生育学分会. 剖宫产瘢痕妊娠诊断与治疗共识[J]. 中华医学杂志, 2012, 92(25): 1731-1733
The family planning association of Chinese Medical Association. The diagnosis and treatment consensus of cesarean scar pregnancy[J]. Natl Med J China, 2012, 92(25): 1731-1733

[10] Jurkovic D. Cesarean scar pregnancy and placenta accreat [J]. Ultrasound Obstet Gynecol, 2014, 43(4): 361-362

[11] Zhang B, Jiang ZB, Huang MS, et al. Uterine artery embolization combined with methotrexate in the treatment of cesarean scar pregnancy: results of acase series and review of the literature [J]. J Vasc Interv Radiol, 2012, 23: 1582-1588

[12] Bayoglu Tekin Y, Mete Ural U, Balik G, et al. Management of cesarean scar pregnancy with suction curettage: a report of four cases and review of the literature [J]. Arch Gynecol Obstet, 2014, Jan 25 [Epub ahead of print]

[13] Yamaguchi M, Honda R, Uchino K, et al. Transvaginal methotrexate inject for the treatment of cesarean scar pregnancy: Efficacy and subsequent fecundity[J]. J Minim Gynecol, 2014, Apr 14[Epub ahead of print]

[14] Uysal F, Uysal A, Adam G. Cesarean scar pregnancy: diagnosis, management, and follow-up[J]. J Ultrasound Med, 2013, 32: 1295-1300

[15] 欧阳振波, 徐玉静, 马云燕, 等. 剖宫产后子宫瘢痕妊娠合并宫内妊娠的诊断及治疗[J]. 现代妇产科进展, 2014, 23(4): 884-887
OuYang Zhen-bo, Xu Yu-jing, Ma Yun-yan, et al. The diagnosis and treatment of heterotopic cesarean scar pregnancy [J]. Prog Obstet Gynecol, 2014, 23(4): 884-887

[16] 何鑫, 陈薇, 张震宇, 等. 子宫动脉化疗栓塞治疗瘢痕妊娠的 Meta 分析[J]. 首都医科大学学报, 2013, 34(2): 275-281
He Xin, Chen Wei, Zhang Zhen-yu, et al. Meta-analysis of the effectiveness of UACE and systematic MTX in the treatment of CSP [J]. Journal of Capital Medical University, 2013, 34(2): 275-281

[17] Kai K, Shimamoto K, Matsumoto H, et al. Conservative surgical treatment for caesarean scar pregnancy [J]. J Obstet Gynaecol, 2014, 34(1): 91-92

[18] 李燕娜, 魏炜, 张军. 腹腔镜在治疗剖宫产后子宫瘢痕妊娠中的应用[J]. 实用妇产科杂志, 2012, 28(4): 285-287
Li Yan-na, Wei Wei, Zhang Jun. Laparoscopy in the Treatment of Cesarean Scar Pregnancy [J]. Journal of Practical Obstetrics and Gynaecology, 2012, 28(4): 285-287

[19] 刘玉昆, 孟丽丽, 刘梅兰, 等. 剖宫产瘢痕妊娠 27 例诊治分析[J]. 中华产科急救电子杂志, 2012, 1(1): 37-41
Liu Yu-kun, Meng Li-li, Liu Mei-lan, et al. cesarean scar pregnancy analysis of 27 cases [J]. Chin J Obstet Emerg (Electrinic Edition), 2012, 1(1): 37-41

[20] Zhang XB, Zhong YC, Chi JC, et al. Caesarean scar pregnancy: treatment with bilateral uterine artery chemoembolization combined with dilation and curettage[J]. J Int Med Res, 2012, 40(5): 1919-1930

（上接第 2208 页）

[18] 朱长虹. 生殖药理学[M]. 北京: 人民卫生出版社, 2007: 154-155
Zhu Chang-hong. Reproductive pharmacology [M]. Beijing: People's medical publishing house, 2007: 154-155

[19] 庄依亮. 子宫收缩抑制剂的药理与临床应用 [J]. 中国新药与临床杂志, 1999, 18(3): 60-62
Zhuang Yi-liang. The pharmacology and clinical apply of Inhibitors of uterine contraction [J]. Chinese Journal of New Drugs and Clinical Remedies, 1999, 18(3): 60-62

[20] 余黎, 王坚. 具有子宫兴奋作用的中药研究概况[J]. 广东药学院学报, 2002, 18(1): 53-55
Yu Li, Wang Jian. Status of uterine stimulant about Chinese medicine [J]. Academic journal of Guangdong college of pharmacy, 2002, 18 (1): 53-55