

# Pharmaceutical Applications and Clinical Efficiency of Anti-Inflammatory Ramon Preparation

Dmitriy Yu. Korulkin and Raissa A. Muzychkina

**Abstract**—The Ramon preparation is received from a plant; it is destined for external treatment of inflammations in post-surgery period. The Ramon is a biogenic immune stimulator accelerating metabolism, contributing to improvement of blood indexes, having general tonic, anti-inflammatory and bactericidal effect.

**Keywords**—Anti-inflammatory, anthraquinones, bactericidal activity, Ramon.

## I. INTRODUCTION

REGARDLESS of comprehensive research and development of new methods of prevention and treatment of suppurative-septic diseases in obstetric-gynecologic practice, the frequency of this pathology is not being reduced and as per data of different authors it is 65%.

The character of microorganisms (their virulence, reproduction temp, contamination degree) and the matter concerning relation of macroorganism and microflora play a big role in development of post-surgery complications. Moreover, many unfavorable factors such as anemia, pyelonephritis, tonsillitis, coleitis, major intraoperative blood loss significantly increase the risk of post-surgery complications, as they disturb protective body functions [1].

In this connection, search and evaluation of new effectiveness of new medicines having biostimulating effect and accelerating regeneration processes that can be used for treatment of post-surgery infectious complications is very promising and topical.

Chemotherapeutical and pharmacological tests as well as experience of folk medicine show that individual natural anthraquinones and their synthetic analogues have versatile physiological activity, and the highest effect was registered for glycosised mono, dimer and reduced forms and for anthracycline antibiotics. Oxidized forms of a glycones exhibit depending on dose astringent and (or) purgative effect, moderate antitumor, anti-inflammatory, antibacterial and radio protective action [2].

Purpose of the study is to prove the effectiveness and safety of the medicine for human with inflammatory infiltrates in obstetric-gynecologic practice.

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## II. RESULTS AND DISCUSSION

Patients with inflammatory infiltration and partial suture line disruption observed pain relieving and wound-healing effect on the second day of using it, wound debridement lasted, as a rule, within the first two days. No hyperemia was observed in the area of sutures on 2<sup>nd</sup> to 3<sup>th</sup> day of using the medicine, and further good dynamics was demonstrated. The absence of clinical effect of the medicine was not observed.

Maternity patients at the age from 19 to 40 years old with clinical signs of complicated wound process after Caesarean operation and perineum: 5 -after Caesarean operation, 7– with complicated wound process at perineum All 100% of patients had complaints on pain in the area of sutures on perineum. Partial suture line disruption on perineum was observed with 3 (42.8%) patients. Patients after Caesarean operation in 2 cases (40%) had suture line disruption as well; in 2 cases sutures were disrupted (40%). In 100% of cases, i.e. all patients had clinical symptoms of inflammation. 9 patients (75%) had light and moderate anemia, 5 (41.6%) had kidney diseases in their disease histories. Pregnancy course of 58.3% was complicated with gestosis, 33.3% - coleitis, sexually transmitted diseases 25% of patients had. Caesarean operation was performed to 4(80%) as urgent procedure, 2 (28.6%) had perineal rupture of first and second degree; episiotomy was performed to 5 (71.4%) patients.

In total blood cell count leukocytosis decrease from 12.5 0.3 10 /l down to 9.7 0.2 10 /l and leukogram normalized on 3-th day from the incursion of disease.

Bacterioscopy of wound content before the treatment was indicative of high contamination with pathological microorganisms (streptococci, staphylococci etc.) in 33.3% of patients, edema, cell hypertrophy and leucocyte content was increased in wound content of all patients. Upon treatment completion it was observed that the number of changed cells reliably decreased, no inflammatory response of epithelium was observed.

During analysis of initial immunogram, the decrease of absolute and relative lymphocyte content down to 16.6±1.2 was observed in the most (77.7%) patient's organism, in 55% immune serum globuline IgM increased (24.8±6.7mg). Immunogram indicators after treatment: reliable increase of lymphocyte ratio to lower values of normal range as well as immune serum globuline IgM normalization was observed [3].

The represented data mean that clinical effect was accompanied with normalization of changed laboratory indicators. We have conducted comparative analysis of Ramon medicine and Levomekol ointment. For this, two groups of patients with clinical signs of inflammation in the

area of post-surgery wounds were examined: main group (37 patients) treated with Ramon medicine and reference group (35 patients) treated with Levomekol ointment.

Comparative evaluation of efficacy of these medicines was performed based on disappearance or reduction of inflammatory infiltrate, edema, wound hyperemia, analysis of frequency and character of side effects and complications, studying parameters of inflammatory blood reaction (leukocytosis, leucogram, erythrocyte sedimentation rate) and data of bacterioscopic analysis of wound content [see Table I-III].

TABLE I  
THE DYNAMIC OF CLINICAL-LABORATORY, INSTRUMENTAL DATA RELATED TO PATIENTS DURING HOSPITALIZATION

Indicators	Main group (37 patients)		Reference group (35 patients)	
	Absolute number	%	Absolute number	%
Skin itching				
- mild	6	16.3	7	20
- moderate	14	37.8	6	17.1
- acute	17	45.9	18	51.4
Edema				
- mild	3	8.1	4	11.4
- moderate	19	51.4	24	68.5
- acute	15	40.5	17	48.5
Hyperemia				
- mild	9	24.3	10	28.5
- moderate	12	32.4	14	40
- acute	16	43.3	16	45.7
Leukocytosis				
- mild	10	27	8	22.8
- moderate	8	21.6	13	37.1
- acute	19	51.4	14	40
Leucogram				
- mild	9	24.3	11	31.4
- moderate	10	27	9	25.7
- acute	18	48.7	15	42.8
ESR				
- mild	9	24.3	7	20
- moderate	10	27	9	25.7
- acute	18	48.6	19	54.3
Bacterioscopic analysis of wound content				
- mild	4	10.8	1	2.8
- moderate	15	40.5	14	40
- acute	18	48.7	20	57.1

TABLE II  
THE DYNAMIC OF CLINICAL-LABORATORY, INSTRUMENTAL DATA RELATED TO PATIENTS ON 5-TH DAY

Indicators	Main group (37 patients)		Reference group (35 patients)	
	Absolute number	%	Absolute number	%
Skin itching				
- mild	27	72.9	22	62.8
- moderate	7	18.9	7	20
- acute	3	8.2	6	17.1
Edema				
- mild	26	70.2	18	51.4
- moderate	9	24.3	11	31.4
- acute	2	5.4	6	17.1
Hyperemia				
- mild	27	72.9	16	45.7
- moderate	8	21.6	12	34.3
- acute	2	5.4	8	22.8
Leukocytosis				
- mild	21	56.7	14	40
- moderate	10	27.0	11	31.4
- acute	6	16.3	10	28.5
Leucogram				
- mild	20	54.0	18	51.4
- moderate	11	29.7	8	22.8
- acute	6	16.3	9	25.7
Erythrocyte sedimentation rate				
- mild	25	67.6	19	54.3
- moderate	8	21.6	10	28.5
- acute	4	10.8	6	17.1
Bacterioscopic analysis of wound content				
- mild	27	72.9	23	65.7
- moderate	7	18.8	8	22.8
- acute	3	8.2	4	11.4
Being satisfied with the medicine used				
- mild	0	0	1	2.8
- moderate	11	29.7	9	25.7
- acute	26	70.3	25	71.4
Character of side effects	No		No	

TABLE III  
THE DYNAMIC OF CLINICAL-LABORATORY, INSTRUMENTAL DATA RELATED  
TO PATIENTS IN 2 WEEKS

Indicators	Main group (37 patients)		Reference group (35 patients)	
	Absolute number	%	Absolute number	%
Skin itching				
- mild	35	94.6	30	85.7
- moderate	2	5.4	4	11.4
- acute	0	0	1	2.8
Edema				
- mild	37	100	29	82.8
- moderate	0	0	5	14.2
- acute	0	0	1	2.8
Hyperemia				
- mild	37	100	31	88.5
- moderate	0	0	3	8.5
- acute	0	0	1	2.8
Leukocytosis				
- mild	37	100	29	82.8
- moderate	0	0	5	14.2
- acute	0	0	1	2.8
Leucogram				
- mild	37	100	29	82.8
- moderate	0	0	5	14.2
- acute	0	0	1	2.8
erythrocytesedimentation rate				
- mild	36	97.3	31	88.5
- moderate	1	2.7	3	8.5
- acute	0	0	1	2.8
Bacterioscopic analysis of wound content				
- mild	37	100	33	94.2
- moderate	0	0	2	5.7
- acute	0	0	1	2.8
Being satisfied with the medicine used				
- mild	0	0	0	0
- moderate	0	0	5	14.2
- acute	37	100	30	85.7
Character of side effects	No		No	

Patients with inflammatory infiltration and partial suture line disruption observed pain relieving and wound-healing effect on the second day of treatment, wound was debrided as a rule within the first 2 days. Also, no hyperemia was observed in the area of sutures on 2 to 3 days of treatment with this medicine, and good dynamic was observed. Identical effects were observed during treatment with Levomekol medicine, but on 3 to 4 day of treatment.

During treatment with Ramon medicine count leukocytosis reduced and leucogram parameters were normalized in full blood cell count on the 3<sup>th</sup> day from the beginning of treatment, while the same clinical effect was observed on 4<sup>th</sup> day of treatment [3], [4].

Bacterioscopic analysis of wound content prior to the treatment gave the evidence of contamination with pathological microflora. High leucocyte count in wound content, edema and cell hypertrophy was observed with all patients.

After treatment with the both medicines it was observed, that the number of changed cells reduced reliably, no

inflammatory reaction of epithelium was observed (see Table IV).

TABLE IV  
COMPARISON OF THE EFFECTIVENESS OF RAMON AND LEVOMEKOL  
MEDICINES

Effects	When effect is demonstrated from the beginning of treatment	
	Ramon (37 patients)	Levomekol (35 patients)
Post-surgery wound debridement	2-th day	3-d day
Edema of post-surgery wound	2-th day	3-d day
Hyperemia in the area of post-surgery sutures	3-d day	4-th day
Leukocytosis reductions	3-d day	4-th day
IgM level normalization	3-d day	4-th day

Thus, the represented data mean that clinical efficacy of Ramon medicine is higher compared to the efficacy of Levomekol ointment. Ramon medicine has high anti-inflammatory effect and it is effective medicine for external treatment of inflammation processes in post-surgery period, it is convenient to use, well-tolerated by patients, has no side effects and can be recommended for wide use in practice.

The Ramon preparation does not contain corticosteroids, hormonal substances etc.

Ramon preparation is allowed for use and for industrial output as a medicine at the Russia and the Republic of Kazakhstan. Ramon is included in the State Register of the most essential for the Republic of Kazakhstan.

### III. EXPERIMENTAL DESIGN

*Prospective research:* Treated (main) group - 37 patients with complicated surgical wound using Ramon & reference group - patients with complicated surgical wounds using Levomekol.

*Methods of research:* General clinical-laboratory data; bacterioscopic analysis of wound content; immune status determination & local visual inspection of wound

*Method of medicine application* is surgical wound treatment 2 times a day; the wounds were not treated using other local means during clinical tests.

Prior to and during treatment all women passed general clinical examination, their immune status was determined, wound content was analyzed using bacterioscopy, medical history data were determined more precisely, the time of wound debridement and healing, forming of full grain tissues, side effects and complications and satisfaction with the medicine taken were estimated.

Ramon drug effectiveness, safety and tolerance was studied based on the disappearance or reduction of inflammatory infiltrate, edema, wound hyperemia, analysis of frequency and character of side effects and complications, studying parameters of blood inflammatory reaction (leukocytosis, leucogram, erythrocytes dimentation rate), immune status and wound content bacterioscopic data [5]-[6].

*The received results were statistically processed* according to the generally accepted medicine statistic methods.

*Characteristic of the analyzed group:* during clinical test of Ramon medicine 37 patients at the age of 35 to 57 with clinical signs of complicated post-surgery process were examined and treated. All patients had complaints on pain in the area of post-surgery sutures. 15 (40.5%) patients had edema in the area of sutures, 16 (43.3%) patients had tissue hyperemia and edema, partial suture line disruption was observed with 6 women (16.2%). Partial suture line disruption observed with 2 patients of 37 ones was accompanied by temperature increase up to 37.7 Celsius degrees, rigor, and general uneasiness. 26 patients (70.3%) had chronic centers of infection - tonsillitis, pyelonephritis, adnexitis in their medical histories.

The blood of all patients was analyzed for leukocytosis, ESR, bacterioscopic smear analysis was performed, some chains of humoral immunity were determined prior and during treatment.

Moreover, 12 maternity patients with clinical signs of inflammation at sutures on anterior abdominal wall after Caesarean operation and partial suture line disruption on perineum were examined.

All patients passed general clinical test, bacterioscopic analysis of wound content, medical history data were taken into account, time of wound debridement and healing and grain forming was taken into account.

*Treatment effectiveness* was analyzed based on the disappearance or reduction of edema and tissue hyperemia, with account of parameters of blood inflammatory reaction, wound content bacterioscopic data analysis [3].

*Ramon medicine:* 2% Ramon ointment manufactured by Santo Member of Polpharma Group (Kazakhstan).

*Drug for comparison:* 5% Levomekol content manufactured by Nizhpharm (Russia).

*Antibacterial assay:* Different concentrations of the Ramon substance were tested for antibacterial activity using agar disc diffusion assay according to the method [3]. The strains obtained were inoculated in conical flask containing 100 ml of nutrient broth. These conical flasks were incubated at 37C for 24h and were referred to as seeded broth. Media were prepared using Muller Hinton Agar (Himedia), dispensed on petri dishes and lawn cultures were prepared using sterile cotton swabs with the test organisms from the seeded broth. Sterile discs of six millimeter width impregnated with 20 µl of test preparation in different concentrations were placed on the upper layer of the lawn cultures. Streptomycin (10µg/disc) was used as standard. The plates were incubated overnight at 37°C.

Antibacterial activity of the Ramon was assayed by measuring the inhibition zone formed around the discs. The experiment was repeated triplets and the mean values were calculated.

*Mutagenous activity* was studied on bacterial strains as nutrient medium reagents ampicillin, DMSO, D-biotin, L-histidin, D-glucose-6-phosphate, NADF, phenobarbital, MPB, M9 medium, 1.5% minimal glucose agar, 0.6% minimal agar were used. Chemical mutagens – N-methyl-N-nitrosomethylcarbamide, 2,7-diamino-4,9-dioxy-4,5,9,10-tetrahydro-

5,9-diozopyren. As the negative control variants with dissolvent were used. Supernatant (C 9-fraction) was prepared according to special method with +4 C degrees immediately before the tests. Tests were held according to Ames B method and others.

The toxic of the Ramon and its components was defined on 9-days hens' embryos with 5 repetitions. The result showed the absence of mutagenous activity [3].

*Microbic dissemination* of 2% ointment was studied with the screen method, with 3 repetitions, into Petri cups with MPA nutrient medium from serial dissolved initial medicinal form and 100 ml of sterile water. The content of inoculation was cultivated in thermostat at 37 C degree during 48 hours with the following calculations of microorganisms' colonies on cups. Microbic dissemination of studied 2% ointment on lanolin base was 4. 5. 10 per 1g.

*Microbiological cleanses of medicinal form* was checked also according to the method of the State Pharmacopoeia of the Republic of Kazakhstan.

Medicinal means was studied in from 2 to 0.00001% concentration with 3 repetitions. It showed that substance with 0.07 to 0.14% concentration inhibits the growth of colibacillus, staphylococcus and pseudomonids, and with 0.016 to 0.047% doses delays the growth of these inoculations. Sporogenic bacteria and yeast inoculation are more resistant to the Ramon, minimal suppressing concentration for them is 0.14 to 0.28%. 2% ointment fully inhibits the growth of all used test-cultures, MPD=0.2%. Medicinal forms on lanolin base with lower than 0.2% of concentration does not have antimicrobial effect.

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