City Clinical Hospital № 24

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Report
on phase II clinical trial of Polyoxidonium in patients with diffused forms of colon carcinoma

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Report

Clinical study of 6 mg Polyoxidonium injections in patients with diffused forms of colon carcinoma

Type of study: Double blind, placebo – controlled

The present research conduction is based on the decision of the Pharmacological Committee of Ministry of Health and Medical Manufacturuing, Russian Federation made on 03.10.1995.

Phase of study: II

Number of patients: 95

Serial number of medication: 030995

Sponsor: "Scientific – Medical Center Petrovaks"

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Regulating standards:
The research is carried out in accordance with the requirements of Pharmacological Committee, Ministry of Health, Russian Federation
The BS project "Regulations of conduction of high-quality clinical studies in Russian Federation"

Contents Pages

1. Grounds for the present research conduction

The decision of the Pharmacological Committee of Ministry of Health and Medical Manufacturing, RF, made on 03.10.95., gave grounds for the II phase clinical study of medication "Polyoxidonium" . According to the Protocol #18 of Pharmacological Committee Meeting held on 3 October 1996, the City Clinical Hospital #24 was approved to be the research of the Polyoxidonium clinical trials.

II. Aims and objectives of the present trial

The present clinical research was primarily aimed to evaluate the effectiveness of different treatment schedules of the medication "Polyoxidonium" in patients with diffused forms of colon carcinoma. In addition, influence of the probationer medication on the immunological status values of the given patients was studied as well.

III. General research plan

1. Patient eligibility criteria

Patients that had undertaken either palliative or conditional-radical operation on account of diffused forms of colon carcinoma were enrolled in the study.

Diagnostic criteria included intraoperative findings histological test results and data retrieved by means of laboratory methods (survey X-ray of the chest, ultrasonography of abdominal cavity and pelvic organs, laparoscopy, computer tomography and s.o.).

Overall status of patients by the moment of therapy commencement was evaluated by treating doctors as to be satisfactory.

All patients participated in the research as volunteers. All patients had received the written information on medication Polyoxidonium and subsequently, they had given the written informed consent to take part in the study.

The patients with critical status, those with evident signs of hepato – renal failure as well as patients suffering from the unsanated surgical infectious processes were excluded from the study.

2. Treatment schedules

The phase II clinical trial bore "double-blind" character, in other words, neither patients nor physicians knew whether patients receive Polyoxidonium or placebo. Research supervisor randomized the patients. Researches had at disposal the numbered and sealedenvelopes that contained special codes. In cases of development of undesirable adverse effects as well as unutilized flasks containing medication were sent back to the developer. The special forms were filled in cases of adverse effect manifestation.

Identical flasks containing either 6 mg Polyoxidonium or placebo produced by the Institute of Immunology were used during research. Each flask was marked with identification number, which corresponded to the patient`s questionnaire number. Medications were kept in a locked refrigerator at +4°C.

The following three injection schedules were used during Polyoxidonium therapy:

I schedule – Patients received either Polyoxidonium (main group) or placebo (control group) given intramuscularly at a single dose 6 mg in the morning for 5 successive days (course dose – 30 mg);

II schedule – intramuscular injections given at single dose 12 mg in the morning of alternate days for 10 days (course dose – 60 mg)

III schedule – intramuscular injections given after the completion of chemotherapy course at single dose 12 mg once a week for 1 month (course dose – 60 mg).

Immunomodulatory treatment other than Polyoxidonium was not administered during the trial.

Parallely to Polyoxidonium therapy or placebo patients undertook combined treatment of diffused forms of colon carcinoma in accordance with the schemes that were approved by the research clinic. The main and control group patients were comparable in respect of stage and localization of oncologic disorder, volume and time of surgical intervention as well as regarding combined treatment regiments (Table 1). All patients received Polyoxidonium injections under the doctor`s supervision.

3. Patient status evaluation criteria

All data retrieved during research were kept accurately using "Patient`s Forms" and all information was, subsequently, analyzed by means of mathematical statistics. The required documentation was filled and the exclusion motives were adequately pointed out if the patient was expelled from the study.

3.1. Clinical values

The following clinical status values of each patients were evaluated daily: body temperature twice a day (mornings and evenings), heart rate, systolic and diastolic blood pressures as well as overall patient status according to the conventional 3-grade scale – "satisfactory", "moderate", "severe".

3.2. Hematological values

The complete blood count and the differential white blood cell count including assessment of hemoglobin level, erythrocyte and leukocyte counts, lymphocyte, immature and mature neutrophils` (young, band and segmented neutrophils), eosinophil, basophil, monocyte percentages as well as the review of cellular morphology following routine staining of the blood smear using Giemsa-Romanovsky stain were taken by all study participants the day before Polyoxidonium treatment commencement.

3.3. Biochemical values

Biochemical blood tests were accomplished by apparatus "Minilab" ("Labsystems").

Total protein, albumin, bilirubin, creatine, blood urea nitrogen levels as well as the activity of transaminases (ALT, AST) were assessed using standard biochemical diagnostic sets, suggested by the firm.

3.4. Immunological values

Immunological status evaluation included flow cytometry studies of subpopulations of peripheral blood lymphocytes (CD3+, CD4+, CD8+, CD16+, CD21+ lymphocytes) using monoclonal antibodies. The functional activity of peripheral blood neutrophils was assessed according to luminole-dependent chemiluminescence reaction by CL3604 apparatus ("Mir-Dialog" Moscow) (spontaneous and zymosane-induced NTB-test and LDCL). The serum immunoglobulin levels were assayed using Manchini method of radial immunodiffusion.

4. Clinical and laboratory data evaluation scheme

Physical examination and laboratory tests to evaluate the patient status were performed the day before the immunomodulatory therapy commencement and days after it`s completion.

IV. The medication safety evaluation criteria

The severity and the rate of systemic as well as local adverse effects (allergic reaction, aggravation of intoxication, development of inflammatory or infiltrative reactions on the injection sites, exacerbation of hematological, biochemical, immunological characteristics of patients) related to combined therapy were primarily evaluated on purposes of Polyoxidonium safety determination.

Those patients who developed adverse effects were excluded from the research group, medication use was discontinued, and treatment of side effects was administered if required. The envelope containing the special code was unsealed and the form of adverse effects was filled.

V. Effectiveness evaluation criteria

1. The criteria of clinical efficacy

The subjective evaluation of patient status by physician as well as by the patient himself served to be the basic criterion for the efficacious application of Polyoxidonium during the combined treatment of patients suffering from the diffused form of colon carcinoma.

Polyoxidonium treatment result was registered to be "good effect" (2 grades) – if the patient had exhibited apparent tendency to overall status improvement, increase in work capacity and appetite as well as if the cancer intoxication had diminished following the Polyoxidonium therapy.

Polyoxidonium "ineffectiveness" was proved (1 grades) – if Polyoxidonium use had not yielded any changes in patient status.

"Negative effect" was determined (0 grade) – in the cases of progressive aggravation of patient status, namely, in the cases of the body temperature rise, cancer intoxication enforcement, occurrence or exacerbation of pain.

2. The laboratory criteria of treatment efficacy assessment

Improvement of hematological, biochemical and immunological values (manifested by the normalization of differential white blood cell count, enhancement of hemoglobin level and erythrocyte number, reduction in bilirubin, blood urea nitrogen, creatinine concentrations to the normal values as well as positive changes in quantitative and functional characteristics of immune system) served to be laboratory criteria for the treatment effectiveness.

VI. Results

1. Clinical results of Polyoxidonium use during combined treatment of diffused forms of colon carcinoma

95 patients who had undergone either palliative of conditionally radical surgical operation for diffused forms of colon carcinoma, were examined and treated further during research. All patients had been suffering from the advanced (III – IV stages) forms of disease. In addition to surgical intervention all patients took chemotherapy course with 5-fluorouracile at a dose of 500mg/m^2 of body surface given by continuous injections intravenously on alternate days (total dose 5-7 g by 4-6 injections). Polyoxidonium therapy was administered accordingly to the pointed schedules in parallel with the latter half of the chemotherapy course. I schedule treatment with Polyoxidonium at the course dose 30 mg was administered to 16 patients and additional 14 patients received placebo. 15 patients were treated with Polyoxidonium schedule II and 10 patients receive placebo. Polyoxidonium and placebo in accordance with schedule III were given to 20 patients each.

1.1. Treatment safety assessment

None of patients suffered from any local or systemic adverse effects related to Polyoxidonium injections. Side effects resultant from the cytostatic agent – 5-fluorouracile at a course dose 5-7g were not observed during combined treatment with chemotherapy and Polyoxidonium either. Moreover, none of patients in placebo groups exhibited adverse effects.

1.2. Clinical evaluation of treatment effectiveness

It seems important to characterize in detail the cohort of patients that were included in the clinical trial directed at the assessment of clinical effectiveness of Polyoxidonium use in oncological patients. Thus, 30 out of 95 patients from the main group suffered from the rectal cancer that has been know to be unfavorable prognostic factor in comparison with the involvement of upper colon. Limited tumor dissemination permitted to diagnose T3 stage tumor in 3 patients merely. The local affection was designated as T4 stage during surgical operation in the rest 92 patients. 45 patients exhibited involvement of regional lymphatic nodes and 40 patients had distant metastasis (33 patients with metastatic affection of liver and 7 – with metastasis in lungs). Thus, all patients that were examined and subsequently treated with Polyoxidonium were attributed to III and IV clinical groups in accordance with WHO

classification. Correspondingly, provided treatment aimed primarily not to cure patients but to prolong their life expectancy and to improve the quality of life. Obviously estimation of the Polyoxidonium – related long-term outcome of patients was not included in the study objectives.

Identical cohort of patients comprised control group (patient data are enumerated in the Table 1). Hence, the assessment of clinical results of treatment with either Polyoxidonium or placebo bore mainly subjective character. Physicians and patients observed and noted changes in patients` status and gave grades to the Polyoxidonium therapy using 3 grade – "good effect", "ineffectiveness" and "negative effect" scale described earlier in details.

All patients from the main group that receive Polyoxidonium, reported overall health status improvement, increase in capacity for work and normalization of appetite. However, according to the opinion of treating doctor, 14 out of 51 patients (27,5%) showed no changes in the overall status. "Negative effect" was not observed in the main group patients.

24 out of 44 patients (54,1%) that received placebo did not exhibit improvement of their overall health status following Polyoxidonium therapy. Like main group patients, "negative effect" was not observed in placebo group by physicians or by patients themselves. However, according the physician`s opinion only 10 (22,7%) out of 44 patients benefited from apparent temporary improvement of status, whereas the rest 34 patients remained stable.

The data expounded earlier is demonstrated in the Figure 1.

1.3. Influence of Polyoxidonium therapy on clinical-hematological values of blood test

Significant dynamic changes in clinical-hematological test findings were not observed in either main cohort or control group patients following therapy (Table 2).

1.4. Influence of Polyoxidonium therapy on the biochemical values of blood test

Considerable dynamic changes in biochemical values of blood test resultant from the influence of Polyoxidonium therapy were not found in either main or control group patients (Table 2).

1.5. Influence of Polyoxidonium therapy on the immune status values

Combined treatment with 5-fluorouracile and Polyoxidonium in accordance with the scheme described earlier resulted in positive changes of immune status values in 44 (87,2%) out of 51 patients. In 6 (11,6%) out of 51 patients considerable dynamic changes in immune status parameters were not achieved and in 1 (1,3%) patient negative effect was observed following treatment. Percentages as well as absolute numbers of peripheral blood lyphocytes bearing CD3+, CD4+, and CD8+ immunophenotype showed most apparent increase. It is remarkable that even patients with significant baseline alterations of immune status findings, achieved normalization of impaired immune parameters following administration of medication in accordance with schedule II – 12mg Polyoxidonium given on alternate days. 99,9% of patients, who received one month long course of immunomodulating therapy with Polyoxidonium, exhibited positive dynamic changes in the contents of lymphocyte subpopulations. Serum levels of immunoglobulins, percentage and absolute number of B-lymphocytes as well as functional activity of peripheral blood neutrophils estimated by

luminole-dependent chemiluminescence test (LDCL) remained constant. Nevertheless, it is noteworthy that LDCL-test values turned out to change in a definite individual way: the high baseline values of LDCL-test decreased and those LDCL-test findings that proved initially low rose following Polyoxidonium therapy. Third group patients treated with Polyoxidonium for a month appeared an exception from the general tendency as the given cohort of patients did not show diminution of initially elevated values of LDCL-test following treatment.

Functional activity of peripheral blood phagocytic cells of 25 main group patients estimated by means of adhesion to plastic and NTB-test as well as by quantification of nonenzymatic cationic proteins and by the determination of myeloperoxidase activity appeared devoid of considerable dynamic changes.

Alterations of immune status parameters seemed to be different in a group of patients, who received placebo in addition to chemotherapy. Pronounced negative dynamic changes in immune status values were observed following chemotherapy in 40 out of 44 patients from the given study group and only the rest 4 participants exhibited relative stability in immune status findings (Figure 2). Absolute numbers and percentages of CD3+, CD4+, CD8+, CD16+ lymphocytes as well as in functional activity of peripheral blood neutrophils assessed by LDCL-test demonstrated maximal decrease whereas the immunoglobulin contents in blood serum escaped significant alterations.

25 out of 44 control group patients underwent evaluation of functional activity of peripheral blood phagocytic cells by abovementioned methods. Twenty-one out of these 25 patients exhibited immune status exacerbation, namely decrease in plastic adhesion, NTB-test values, quantity of non-enzymatic cationic proteins and myeloperoxidase activity. Profundity and the specter of these impairments were extremely individual, though indubitable.

The data expounded earlier is enumerated in table 3.

VII Conclusion

The retrieved data permit to suggest that Polyoxidonium inclusion in the combined treatment of oncological patients (in the given case – patients with diffused forms of colon carcinoma) proved to be totally safe.

None of either local or systemic adverse effects or allergic reactions related to Polyoxidonium therapy was observed in the study participants.

Moderate doses of cytostatic preparations that did not generate profound clinical adverse effects used during the present research do not permit us to support the probability of Polyoxidonium use on purpose of prevention from the adverse effects of chemotherapy. Nevertheless, 100% of patients treated with Polyoxidonium reported subjective positive effect following therapy, whereas only 42,9% of control group patients gave approving appraisal. These results might be considered to provide the biased evidence of the medication capability to decrease certain adverse effects (nausea, heartburn, fatigue, irregular stool and s.o.) of cytostatic agents.

The most interesting results were obtained on dynamic evaluation of immune status parameters of study participants. Thus, chemotherapy produced the diminution of practically

all immunological test findings in a prevalent majority of control group patients, whereas the Polyoxidonium use not only contributed to the stability of immune status values but also even yielded increase at certain level in a majority of patients. Considerable improvement in immunocompetent cell functional capabilities that have been shown to be most sensitive to cytostatic influence provide evidence for the capacity of Polyoxidonium to immunomodulate and moreover, to stimulate immunity regardless of immunosuppressive therapy administered in parallel with the probationer medication.

Administration of different treatment schedules yielded decision that Polyoxidonium ese at a course dose 30 mg might appear sufficient for maintenance of normal baseline values of immune status. However, Polyoxidonium application at 60mg course dose in conjugation with chemotherapy proved reasonable in patients with low baseline findings of immunity. Weekly injections of single dose Polyoxidonium are indicated to patients, who develop immunodeficiency during the intervals between the chemotherapy courses. Medication capability to take effect on the phagocyting branch of immune system might turn out to be advantageous to the prevention from the intensive chemotherapy related infectious complications. Moreover, good tolerance of Polyoxidonium and total absence of adverse effects render it`s use perspective in this field of medical practice.

The category and characteristic of the primary disease of patients included in the present research (III and IV clinical group), unfortunately, preclude from the authentical follow-up of long–term outcome of oncological patients treated with Polyoxidonium. However, Polyoxidonium administration in parallel with immunochemotherapy to patients that have already underwent radical operation for oncological disease might appear to be perspective.

Thus, resuming the data expounded earlier one may note that Polyoxidonium application on purposes of prevention and correction of secondary immune deficiency states in oncological patients (on this occasion – in patients with diffused forms of colon carcinoma) is safe and do not lead to the highly effective use of medication in conjunction with different stages and various schemes of chemotherapy.

The packaging of the medication Polyoxidonium – 6mg per flask – that was suggested for the present clinical trial has proved optimal as it enables to administer preparation at a single dose of 6 mg as well as 12 mg (2 flasks).

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Immunologist, MD E.V. Marchuk

Table 1.

Patient distribution in accordance with gender, age, localization and stage of oncological process, volume and time of surgical operation accomplishment

	Placebo	Polyoxidonium
Males	28 (63,6%)	30 (58,8%)
Females	16 (36,4%)	21 (41,2%)
Age	57,6 ± 2,11 (44-72)	59,3 ± 1,97 (45-69)
Height	159,9 ± 2,2 (168-178)	165,4 ± 1,68 (157-180)
L o c a l i z a t i o n		
Appendix	5 (11,1%)	5 (9,8%)
Ascendant colon		
Descendant colon	4 (9,1%)	4 (7,84%)
C. sigmae	5 (11,4%)	8 (15,7%)
Rectum	30 (68,4%)	34 (66,75%)
S t a g e		
T3	7 (14,3%)	3 (5,9%)
T4	37 (85,7%)	48 (94,1%)
N1	11 (25%)	2 (3,92%)
N2	30 (75%)	49 (96,08%)
M0	10 (23,4%)	14 (27,5%)
M1	34 (78,6%)	37 (72,55)
Volume of surgical operation		
Hemicolectomy	12 (29,6%)	12 (25%)
Resection of c. sigmae		3 (6,25%)
Anterior resection	16 (35,7%)	12 (25%)
Abdominal-anal resection	16 (35,7%)	24 (46,7%)
Of them		
Combined	29 (65,5%)	23 (45,7%)
Expanded	15 (34,5%)	28 (54,3%)
Time after the operation (months)	3,64 ± 0,7	3,72 ± 0,56

Table 2.

Dynamic alterations of hematological and biochemical values in the main and control groups

	P l a c e b o		**P o l y o x i d o n i u m**	
	Before	**After**	**Before**	**After**
Leukocytes (billion/l)	7,4 ± 1,3	7,4 ± 1,2	8,0 2,1	7,9 1,4
Neutrophils-				
Band (%)	3,6 ± 0,2	3,4 ± 0,2	3,7 ± 0,14	3,5 ± 0,2
Absolute (billion/l)	313 ± 81	351 ± 86,2	342,5 ± 74,2	299,2 ± 89
Neutrophils-				
Segmented (%)	55,4 ± 10,2	55,8 ± 10,4	50,5 ± 9,8	56,5 ± 10
Absolute (billion/l)	4500 ± 756,2	4987 ± 897	4978 ± 923,2	5121 ± 758
Lymphocytes-				
(%)	26,5 ± 5,2	25,4 ± 3,8	29,5 ± 5,1	32,5 ± 4,2
Absolute (billion/l)	2150 ± 348	1875 ± 457	2289 ± 598	2345 ± 481
Monocytes-				
(%)	6,5 ± 2,3	6,1 ± 1,8	5,7 ± 2,8	6,1 ± 1,8
Absolute (billion/l)	589 ± 98,5	512,2 ± 95,9	516,5 ± 75,5	598,6 ± 80
Hemoglobin (g/l)	108,2 ± 10,2	111,2 ± 9,8	106,6 ± 4,77	104,7 ± 5,8
Total protein (g/l)	78,5 ± 5,8	72,7 ± 6,9	72,7 ± 8,3	74,4 ± 5,5
Albumin (g/l)	39,5 ± 4,8	35,8 ± 4,6	37,2 ± 5,6	37,7 ± 5,2
Bilirubin (mmol/l)	7,6 ± 2,8	7,9 ± 2,5	4,5 ± 1,2	5,5 ± 2,1
BUN (mmol/l)	5,1 ± 0,8	4,8 ± 0,4	4,9 ± 1,1	4,9 ± 1,2
Creatinine (mcmol/l/min)	95,2 ± 12,1	87,8 ± 10,5	77,7 ± 13,5	75,2 ± 12,1
AST (mcmol/min)	19,1 ± 3,5	17,4 ± 4,4	22,4 ± 3,1	23,3 ± 2,9
ALT (mcmol/l/min	12,1 ± 2,1	13,4 ± 2,3	10,4 ± 2,7	11,6 ± 3,1

Table 3

Dynamic changes in immune status findings of study patients

	P l a c e b o (44)		**Polyoxidonium (51)**	
	Before	**After**	**Before**	**After**
CD3+ (%)	68,1 ± 2,9	49,3 ± 3,1*	65,4 ± 3,7	70,0 ± 4,05&
CD3+ absolute (mln/l)	1518,1 ± 219,3	958,8 ± 112,5*	1449 ± 154	1936,8 ± 234,3&
CD4+ (%)	30,6 ± 2,0	23,5 ± 1,7*&	27,8 ± 1,7	36,6 ± 2,2*&
CD4+ absolute (mln/l)	629,9 ± 45,8	429,8 ± 64,1*&	582,1 ± 54,4	934,1 ± 128,6*&
CD8+ (%)	25,7 ± 1,4	23,8 ± 1,54	27,9 ± 1,8	26,6 ± 1,6
CD8+ absolute (mln/l)	565,5 ± 49,9	418,7 ± 37,9*&	571,7 ± 53,3	706,8 ± 74,9
CD4/CD8	1,11 ± 0,1	0,98 ± 0,08&	1,04 ± 0,08	1,46 ± 0,09*&
B-lymphocytes (%)	5,21 ± 2,1	6,14 ± 1,9	4,97 ± 1,33	6,83 ± 2,2
B-lymphocytes absolute (mln-l)	215,5 ± 81,5	231,5 ± 65,8	152,1 ± 58,4	212,1 ± 90,5
IgG (mg%)	1251,6 ± 141,8	1321,8 ± 135,5	1423,13 ± 97,7	1447,5 ± 102,3
IgA (mg%)	357,8 ± 54,8	300,4 ± 78,8	446,9 ± 80,3	412,6 ± 72,3
IgM (mg%)	156,8 ± 57,7	139,9 ± 86,6	199,7 ± 39,0	222,8 ± 43,9
LDCL spont (imp/sec)	589,9 ± 119,8	356,8 ± 194,5	475,2 ± 140,8	826,9 ± 298,6
LDCL ind (imp/sec)	5996,5 ± 986,8	2548,5 ± 568,5*	6991,0 ± 2101,3	5654,1 ± 694,9&
Adhesion (%)	47,5 ± 4,5	31,1 ± 2,8*&	49,6 ± 5,9	51,6 ± 5,7&
NTB-test (c.u.)	145,5 ± 14,7	115,5 ± 12,4&	157,4 ± 18,6	163,5 ± 24,3&
Cationic proteins (c.u.)	56,6 ± 6,1	44,1 ± 5,2&	49,9 ± 6,9	61,4 ± 2,9&
Myeloperoxidase activity (c.u.)	458,9 ± 96,2	347,5 ± 75,5	467,5 ± 86,6	481,4 ± 87,9

The findings marked with * showed statistically significant differences within the group.
Dynamic changes in parameters marked with & showed statistical difference between groups.

Figure 1. Overall health status of patients received placebo and polyoxidonium (PO) therapy (%)

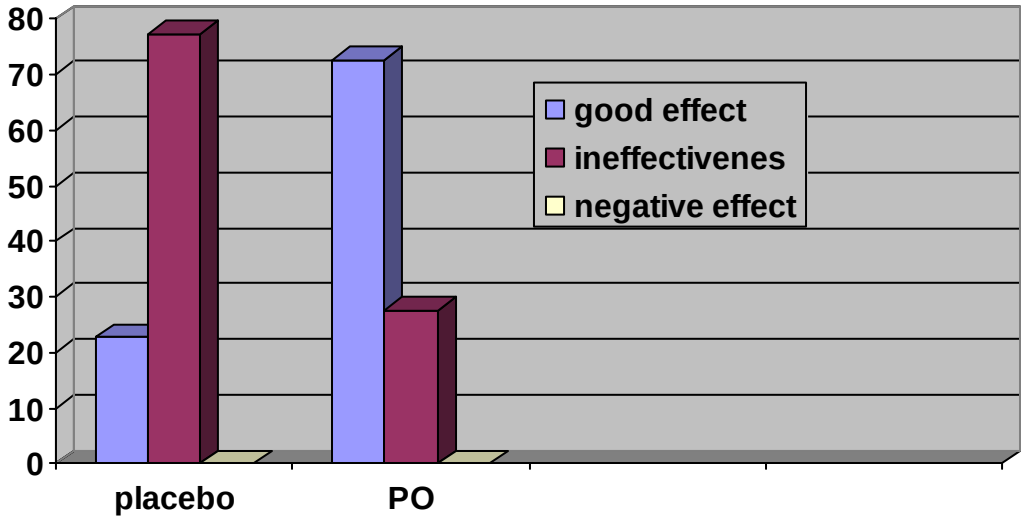

Figure 2. Alterations of immune status parameters (%) in a group of patients