

Immuno.pure[®]

Cytapheresis Adsorber



NIKKISO

Immuno.pure[®]

Cytapheresis Adsorber

el : Συσκευή προαπόφραξης για κυτταραφαίρεση
fr : Siftaferez Citaz
fr : Adsorbent pour cytaférese
es : Adsorbent de citaféresis
pl : Adsorbent do cytoferazy
da : Celleaferease-adsorber

it : Soltafereseadsorberi
pt : Dispositivo de Adsorção de Citafereza

REF CP-MM

LOT

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STERILE R
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Nikkiso provides UC patients with the NEW TREATMENT OPTION “Immunopure” for their QOL improvement.

Current treatments for UC induction of remission

In active ulcerative colitis (UC) refractory to 5-aminosalicylic acid such as mesalazine, either steroids or immunosuppressants/immunomodulators are commonly chosen as alternatives. Both treatments confer considerable risks. Today, many biologics are newly developed and used per escalation of patients' severity.

Adverse events in use of drugs for UC

When steroids and other drugs are prescribed for a long time, patients can be intolerant to them. Severe side effects can occur in drug treatments. Many patients do not prefer to take steroids in the long-term treatment, as they may cause some of adverse events.

Nikkiso proposes cytapheeresis with Immunopure to UC patients

Active UC patients unable to be medicated require other treatments. Cytapheresis is a treatment intended for adsorbing activated blood cells which causes colonic inflammation.

An adsorptive cytapheeresis device such as “Immunopure provided by Nikkiso” is used for UC patients.

Some cases showed that Immunopure could be a promising option for UC treatment. Compared with standard steroids medication such as prednisolone, it was revealed that cytapheeresis showed the similar therapeutic efficacy and safety.¹⁾ Furthermore, cytapheeresis using Immunopure can be the chance to treat without the undesirable side effects of systemic steroids.

References:

1) Kruis, W; Nguyen, P; Morgenstern, J. Novel Leukocyte/Thrombocyte Apheresis for Induction of Steroid-Free Remission in Ulcerative Colitis: A Controlled Randomized Pilot Study. *Journal of Crohn's and Colitis*, 2019, 13(7), 949-953.



Ulcerative colitis and cytapheresis

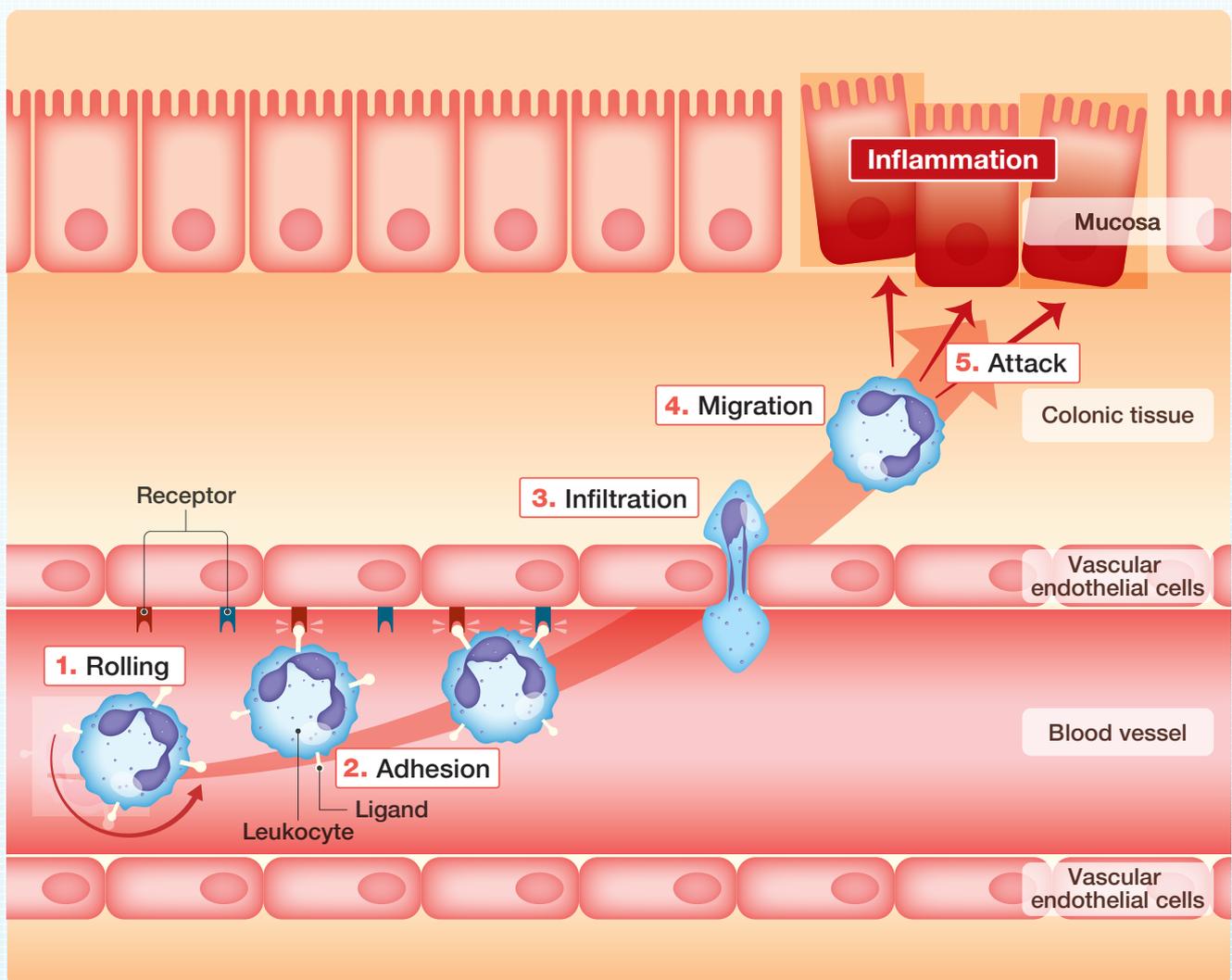
What is ulcerative colitis?

Ulcerative colitis (UC) is an inflammatory bowel disease (IBD) that causes sore and ulceration on mucous membranes of the colon caused by auto-immune systems. UC is a chronic disease, with cycles of active (flare-ups or relapses) and inactive (remission) periods.

In UC, the lining of your rectum and colon becomes inflamed and develops erosions or ulcers. The common symptoms are bloody diarrhea or urgent bowel movements, abdominal cramping, anemia and weight loss.

UC can develop at any ages, but usually starts between the age of 15 to 30. The severity of symptoms varies, depending on the amount of inflammation and its location.

Mechanism of inflammation

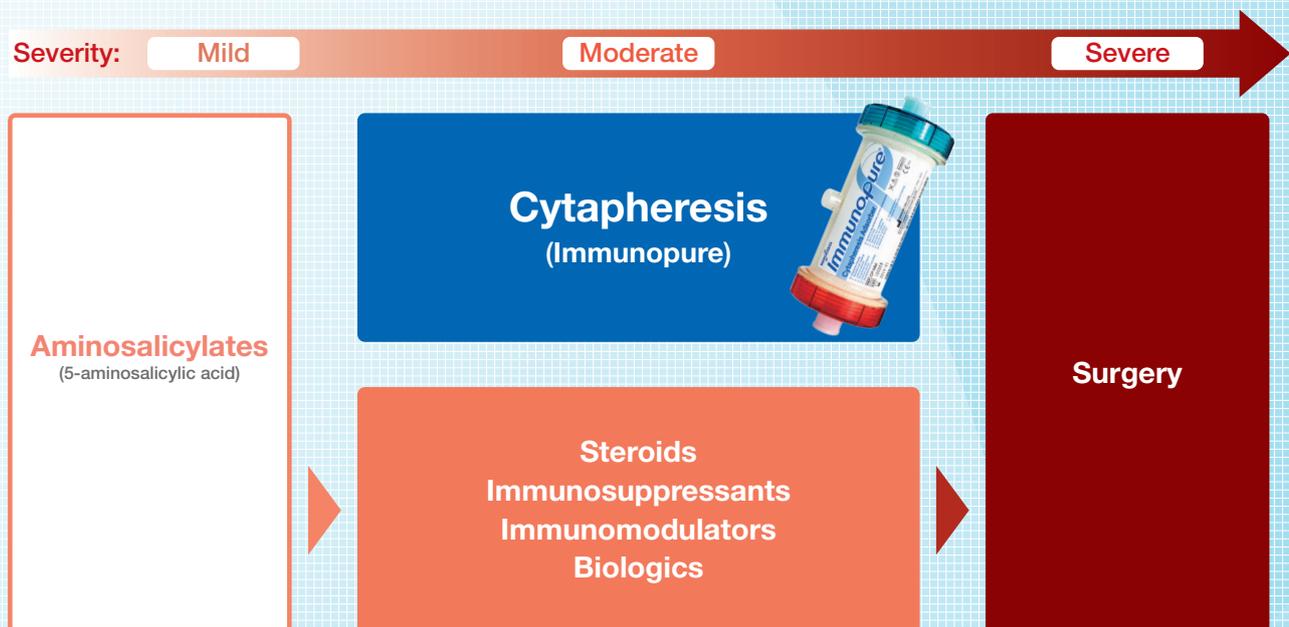


It is suggested that activated leukocytes are recruited around the colonic tissue, attack it and thereby cause inflammation in the colonic mucosa.

What is cytappheresis?

Immunopure is a device to adsorb leukocytes, and is used for cytappheresis. Cytapheresis is a therapy to remove excessive leukocytes and platelets from patients' blood. Activated leukocytes are desirable to be removed as they are considered as one of the obstructive factors in recovering from long-term inflammation of the colon.

■ Induction of remission



Treatments for UC are mainly medicines, cytappheresis or surgery. Among them, medication is often the first choice. There are a variety of drugs available, depending on the severities as follows: aminosalicylates, steroids, immunosuppressants/immunomodulators and biologics.¹⁾ On the other side, their efficacy varies among patients and side effects or intolerance may occur.

Cytapheresis can be applied to patients with steroid-naïve, steroid-refractory or steroid-dependent UC, and in parallel to pharmaceutical treatments.

References:

1) Harbord, M.; Eliakim, R.; Bettenworth, D. et al. Third European Evidence-based Consensus on Diagnosis and Management of Ulcerative Colitis. Part 2: Current Management. *Journal of Crohn's and Colitis*. 2017, 769-784.

What is *Immunopure*?

Simple treatment

Immunopure

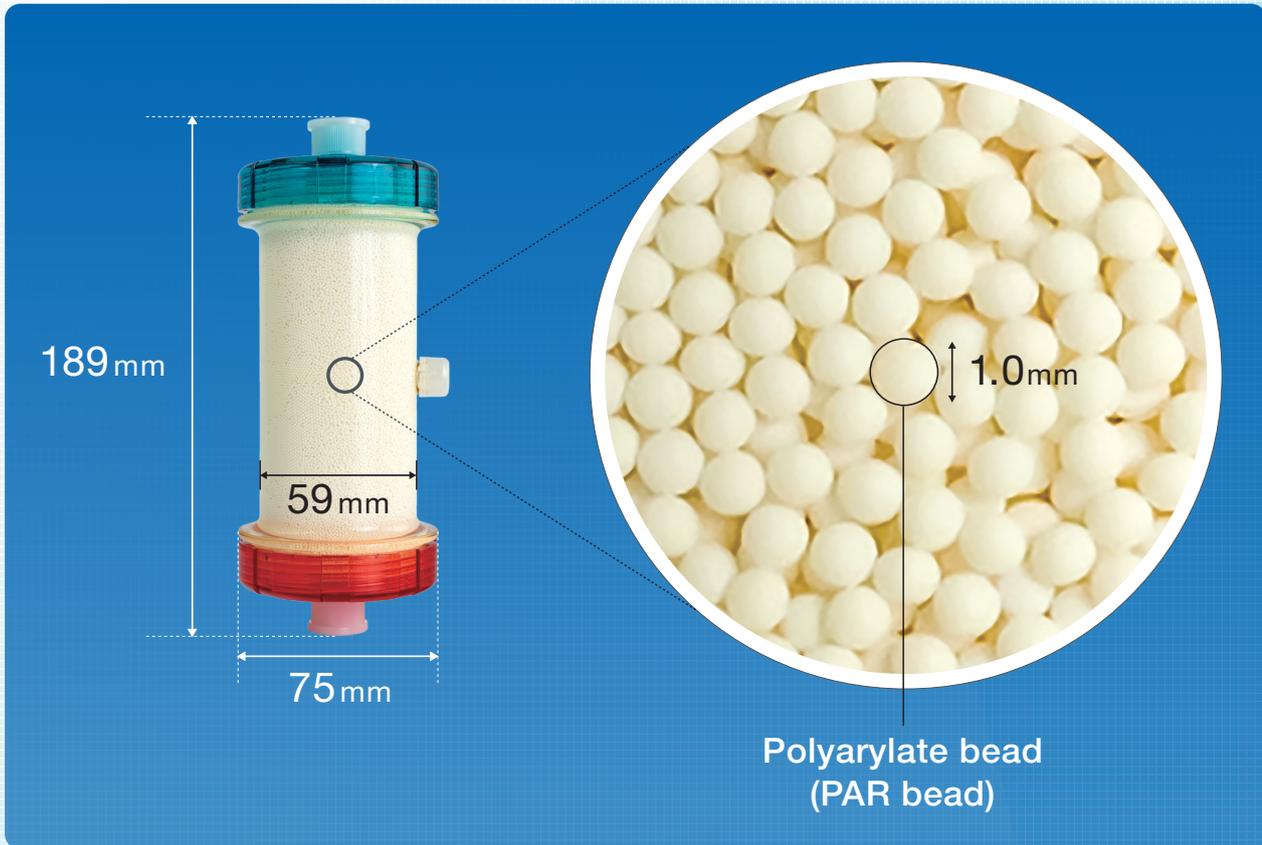
- ▶ is an adsorptive “cytapheresis” device for ulcerative colitis (UC) therapy.
- ▶ is utilized in simple procedure “DHP (direct hemoperfusion) system”.
- ▶ allows selective removal of activated leukocytes.



References:

1) Waitz, G.; Prophet, H.; Ramlow, W. “New White Blood Cell Adsorbent: Immunopure”. Hemoperfusion, Plasmapheresis and Other Clinical Uses of General, Biospecific, Immuno and Leucocyte Adsorbents. Chang, T. M. S.; Endo, Y.; Nikolaev, V. G. et al., eds. World Scientific, 2017, 957-998.

Specifications



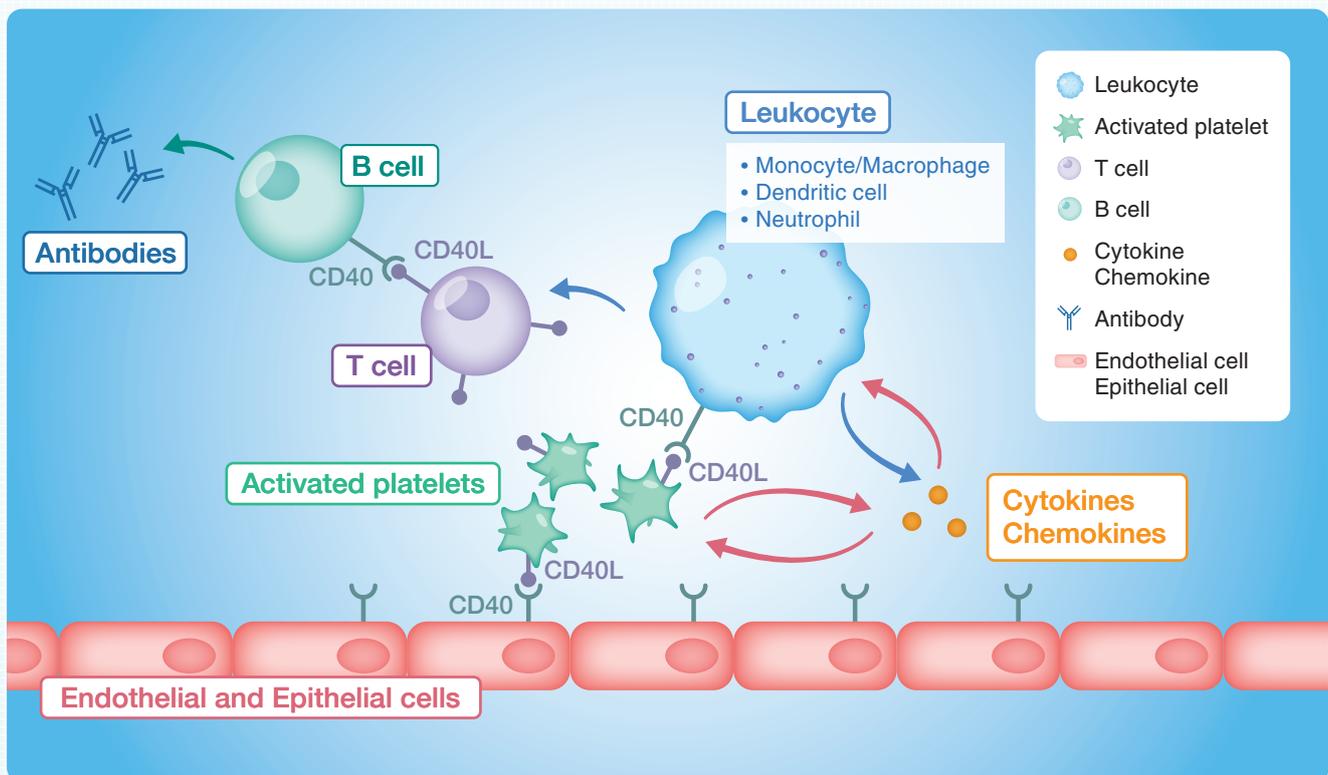
Treatment

| | | | |
|------|------------------|--------|---------------|
| Mode | Circulation rate | Time | Anticoagulant |
| DHP | 30 mL/min | 1 hour | Heparin |

Adsorption mechanism of *Immunopure* 1

Increase of platelet-leukocyte aggregates (PLAs) is observed in UC patients, which plays an important role in the disease.

Figure 1 Platelets activate leukocytes through CD40/CD40L system.



Activated platelets form cytokines and activate leukocytes (monocytes/macrophages, dendritic cells and neutrophils) through CD40L pathway. (Figure 1, red arrows)

Also, B cells activated by leukocytes form antibodies through the pathway. Leukocytes then produce cytokines, chemokines, reactive oxygens species, adhesion molecules and so on. (Figure 1, blue arrows)

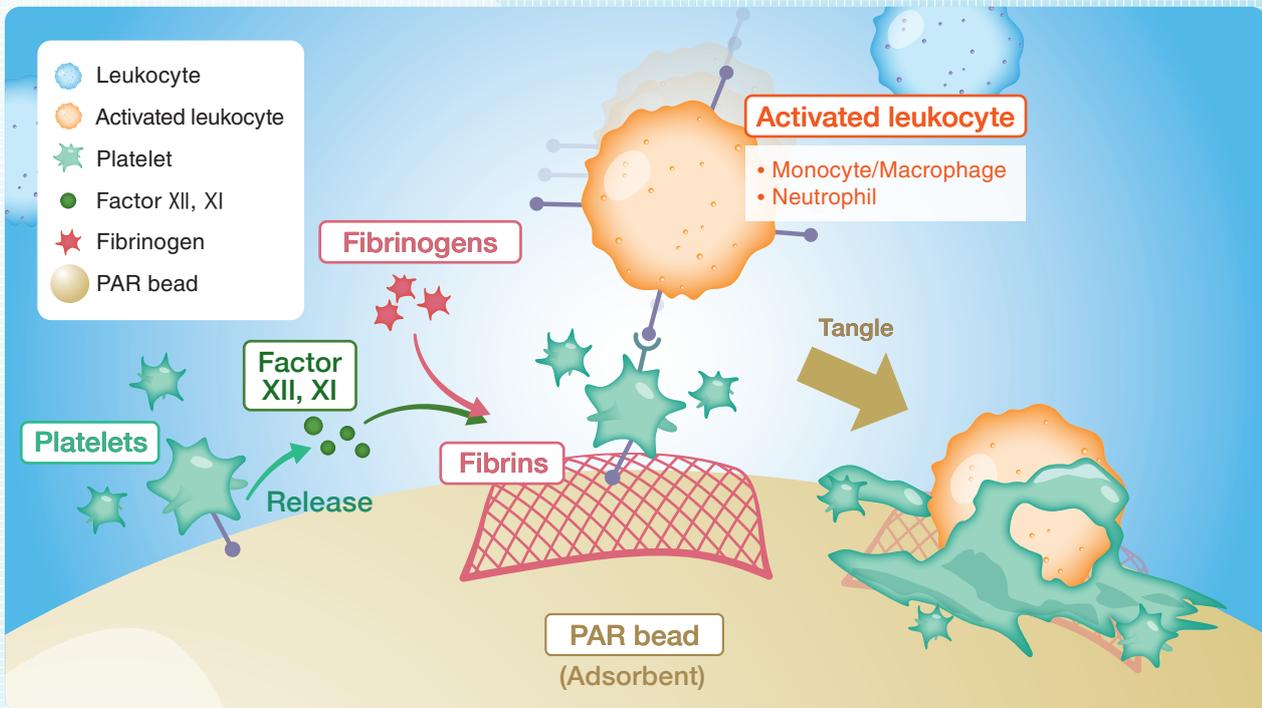
Platelets and leukocytes bind with each other and form platelet-leukocyte aggregates, PLAs. ^{1,2)}

PLAs formed by leukocytes and platelets can lead to severe inflammatory in UC. Increasing number of PLAs were found in the mesenteric vasculature in patients with UC, but not in controls. It is suggested that the activated vascular endothelium stimulates PLA formation in UC. ^{1,3)}

References:

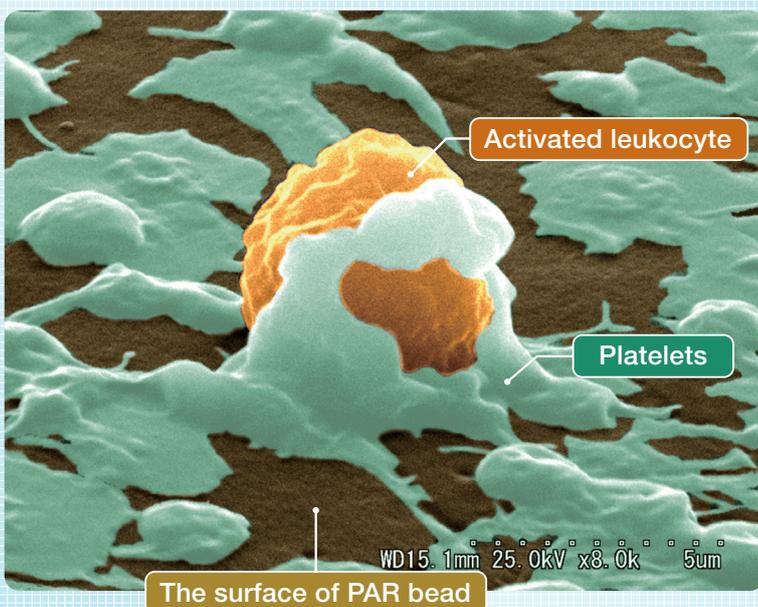
- 1) Waitz, G.; Prophet, H.; Ramlow, W. "New White Blood Cell Adsorbent: Immunopure". Hemoperfusion, Plasmaperfusion and Other Clinical Uses of General, Biospecific, Immuno and Leucocyte Adsorbents. Chang, T. M. S.; Endo, Y.; Nikolaev, V. G. et al., eds. World Scientific, 2017, 957-998.
- 2) Danese, S.; Sans, M.; Fiocchi, C. The CD40/CD40L Costimulatory Pathway in Inflammatory Bowel Disease. Gut. 2004, 53, 1035-1043.
- 3) Pamuk, G. E.; Vural, O.; Turgut, B. et al. Increased Circulating Platelet-Neutrophil, Platelet-Monocyte Complexes, and Platelet Activation in Patients with Ulcerative Colitis: A Comparative Study. Am J Hematol. 2006, 81, 753-759.

Figure 2 The inferred mechanism of tangling of platelets and leukocytes onto PAR bead



It is inferred that when platelets attach to fibrins on PAR bead, an activated leukocyte and platelets are tangled with each other and become an aggregate.¹⁾

Figure 3 The surface of PAR bead observed by electron microscope¹⁾



Aggregates formed by an activated leukocyte and platelets were adsorbed onto the surface of PAR bead.

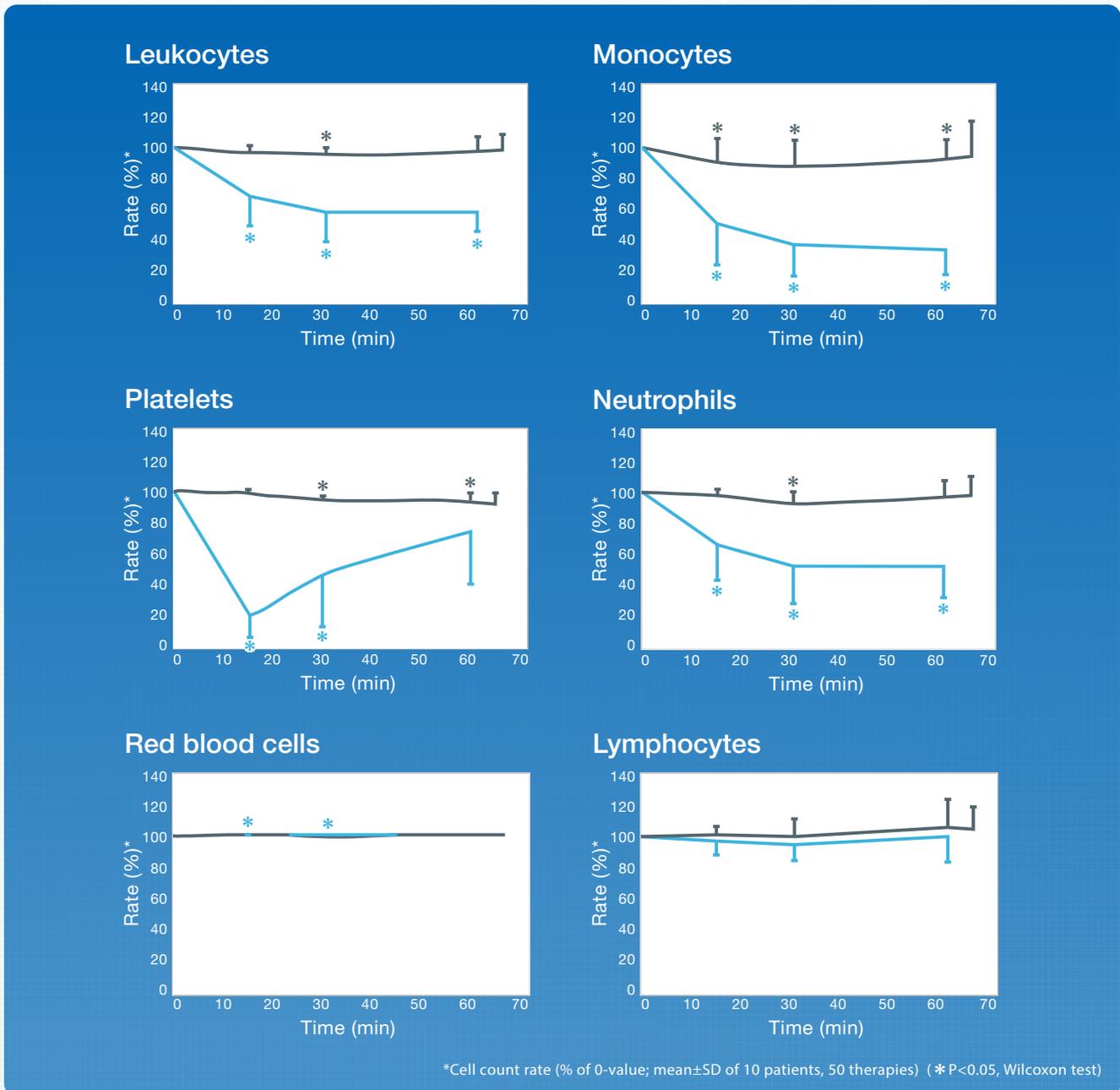
References:

1) Waitz, G.; Prophet, H.; Ramlow, W. "New White Blood Cell Adsorbent: Immunopure". Hemoperfusion, Plasmapheresis and Other Clinical Uses of General, Biospecific, Immuno and Leucocyte Adsorbents. Chang, T. M. S.; Endo, Y.; Nikolaev, V. G. et al., eds. World Scientific, 2017, 957-998.

Adsorption mechanism of *Immunopure* 2

Immunopure can adsorb platelet-leukocyte aggregates (PLAs) formed by activated monocytes, macrophages and platelets.

Figure 1 Removal rate of blood cells ¹⁾ — Inflow — Outflow

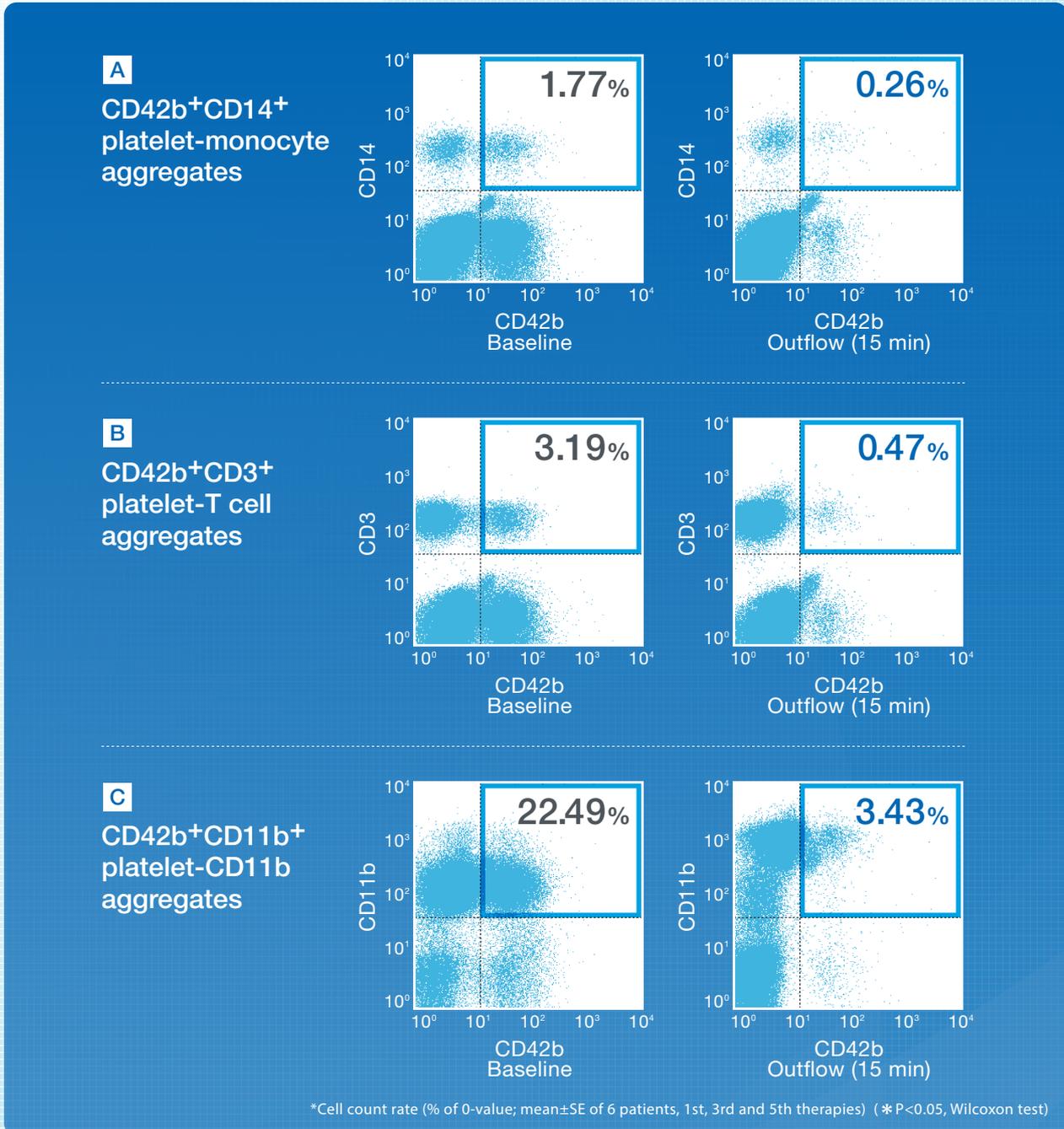


In treatments, leukocytes (monocytes, neutrophils) and platelets were effectively removed while lymphocytes showed little changes. The number of leukocytes was almost stable during the treatment because naïve leukocytes compensated for removed ones. ²⁾

References:

- 1) Ramlow, W.; Waitz, G.; Sparmann, G. et al. First Human Application of a Novel Adsorptive-Type Cytapheresis Module in Patients With Active Ulcerative Colitis: A Pilot Study. *Ther Apher Dial.* 2013, 17(3), 339-347.
- 2) Suzuki, Y. Cytapheresis Conducted on Ulcerative Colitis. *The Journal of the Japanese Society of Internal Medicine.* 2009, 98(1), 61-67.

Figure 2 Flow cytometry analysis of PLAs with CD42b platelet marker ¹⁾



PLAs, formed by platelets and each of CD14⁺ monocytes (**A**), CD3⁺ T cells (**B**) and CD11b⁺ cells (**C**) were counted by flow cytometry analysis.

Ratios of A, B and C reduced after 15-min treatments by the adsorption onto the beads (**A** 1.77%→0.26%, **B** 3.19%→0.47%, **C** 22.49%→3.43%).

PLAs count rates after 15-min treatment in the outflow line compared with 0-value also decreased to 29, 30 or 29% respectively.

References:

1) Waitz, G.; Prophet, H.; Ramlow, W. "New White Blood Cell Adsorbent: Immunopure". Hemoperfusion, Plasmapheresis and Other Clinical Uses of General, Biospecific, Immuno and Leucocyte Adsorbents. Chang, T. M. S.; Endo, Y.; Nikolaev, V. G. et al., eds. World Scientific, 2017, 957-998.

First clinical study in Europe



Study design

Patients Moderate to severe active UC patients : 10 cases
(not exceeding doses of 30 mg prednisolone/day)

In this prospective study, 10 patients (6 males, 4 females, mean age: 47.1 years, between 25 and 73 years) with moderately to severely active UC defined by Rachmilewitz Index (Clinical Activity Index, CAI 6-10) were recruited. They had failed to achieve long-term remission with steroids and/or immunosuppressive agents or had contraindications or were intolerant to high doses of steroids and/or immunosuppressant agents.

Sixty percent of the patients exhibited left-sided colitis (distal colitis) and 40% pancolitis. Their medications comprised 5-aminosalicylic acid, azathioprine, tacrolimus and steroids. Mucosal damage was investigated by Rachmilewitz Endoscopic Index (EI).

Table 1 Protocol ¹⁾

| Time | W1 | W2 | W3 | W4 | W5 | W6 | W7 | W8 | W9 | W10 |
|--------------------|---|---|---|---|---|--|----|----|----|-----|
| Immunopure |  |  |  |  |  |  | | | | |
| Blood tests | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | | | | ✓ |
| Vital tests | ✓ | ✓ | ✓ | ✓ | ✓ | | | | | |
| CAI (Rachmilewitz) | | | | | | ✓ | | | | ✓ |
| EI (Rachmilewitz) | | | | | | | | | | ✓ |

References:

1) Ramlow, W.; Waitz, G.; Sparmann, G. et al. First Human Application of a Novel Adsorptive-Type Cytopheresis Module in Patients With Active Ulcerative Colitis: A Pilot Study. *Ther Apher Dial.* 2013, 17(3), 339-347.

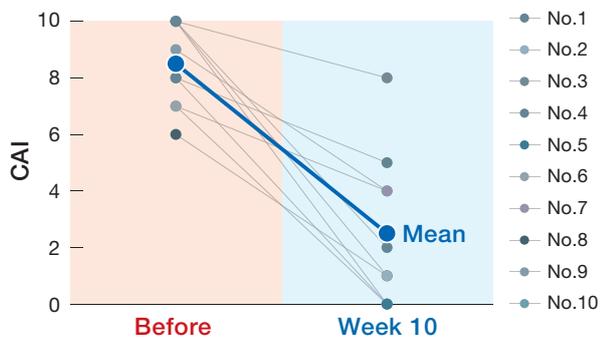


Clinical efficacy

Table 2 Patient data ¹⁾

| Patient No. | CAI Before | CAI W10 | EI Before | EI W10 | Final results |
|-------------|------------|---------|-----------|--------|---------------|
| 1 | 10 | 2 | 6 | 2 | Remission |
| 2 | 10 | 1 | 6 | 2 | Remission |
| 3 | 10 | 8 | 7 | 7 | Non-responder |
| 4 | 8 | 5 | 8 | 10 | Responder |
| 5 | 10 | 0 | 7 | 6 | Remission |
| 6 | 7 | 0 | - | - | Remission |
| 7 | 7 | 4 | 10 | 8 | Remission |
| 8 | 6 | 1 | 7 | 2 | Remission |
| 9 | 9 | 4 | 10 | 8 | Remission |
| 10 | 8 | 0 | 6 | 2 | Remission |
| Mean | 8.5 | 2.5 | 7.4 | 5.2 | |

Figure 1 Rachmilewitz Index ¹⁾



Clinical efficacy was evaluated by Rachmilewitz Index (Clinical Activity Index, CAI).

Clinical remission and response are defined as followed:

- Remission, CAI ≤ 4
- Response, CAI drop ≥ 3 or CAI ≤ 4

In week 10, 8 of the 10 (80%) patients were in clinical remission, 9 of the 10 (90%) were clinical responder.

As a result of Rachmilewitz Endoscopic Index (EI) test, 4 of the 9 (44%) patients were in clinical remission, defined as EI < 4.



Adverse events

Adverse events There was no event in all the 10 cases.

Vital signs No negative effect on vital signs was seen in the study.

References:

1) Ramlow, W.; Waitz, G.; Sparmann, G. et al. First Human Application of a Novel Adsorptive-Type Cytapheresis Module in Patients With Active Ulcerative Colitis: A Pilot Study. *Ther Apher Dial.* 2013, 17(3), 339-347.

ICOP Study in Europe



Study design

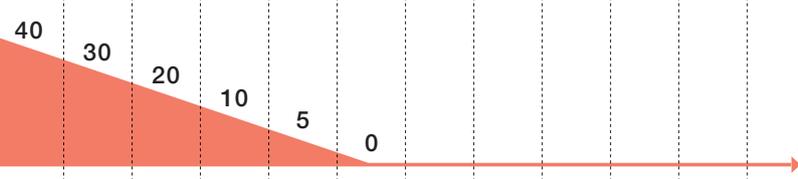
Patients Active UC patients : 22 cases
(refractory to mesalazine)

This study was a controlled, randomized, open study that assessed the efficacy and safety of Immunopure treatment in comparison to prednisolone in active UC.

In this study, 22 patients (12 Immunopures/10 Prednisolones, female:male = 6:6/5:5, mean age: 38.8/38.6 years, between 18 and 75 years) with active UC defined by the Disease Activity Index (DAI 4-8, according to Mayo score) and other scores, despite 5-aminosalicylic acid treatment with ≥ 3 g/day for ≥ 2 weeks, fulfilled the inclusion criteria.

Eighty percent of group Prednisolone were diagnosed with left-sided colitis, 10% with pancolitis and the rest with proctitis, while 66.7% of group Immunopure exhibited left-sided colitis and 33.3% pancolitis.

Table 1 Protocol ¹⁾

| Time | W1 | W2 | W3 | W4 | W5 | W6 | W7 | W8 | W9 | W10 | W11 | W12 |
|-----------------------|---|---|---|---|---|--|--|----|----|-----|-----|-----|
| Immunopure |  |  |  |  |  |  | | | | | | |
| Prednisolone (mg/day) | 40 | 30 | 20 | 10 | 5 | 0 |  | | | | | |
| CRP | ✓ | | | | | | | | | | | ✓ |
| DAI (Mayo Score) | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | | | | | | ✓ |
| Vital tests | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | | | | | | ✓ |

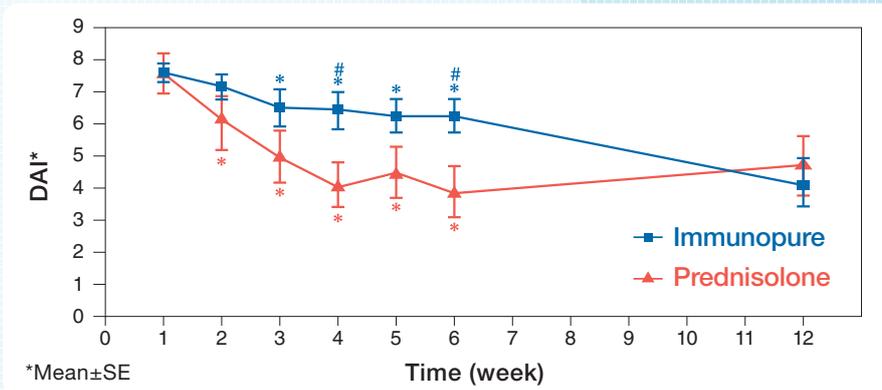
References:

1) Kruis, W; Nguyen, P; Morgenstern, J. Novel Leukocyte/Thrombocyte Apheresis for Induction of Steroid-Free Remission in Ulcerative Colitis: A Controlled Randomized Pilot Study. Journal of Crohn's and Colitis, 2019, 13(7), 949-953.



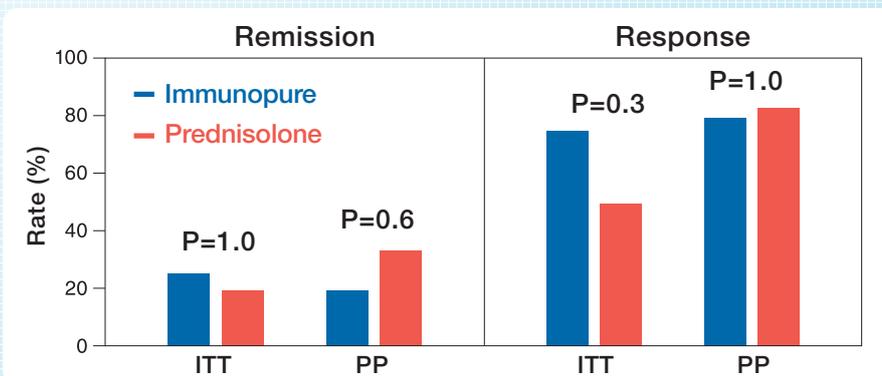
Clinical efficacy

Figure 1 Mayo Score ¹⁾



In the early stage DAI of group Immunopure was higher than group Prednisolone, while it decreased to the same extent in week 12.

Figure 2 Remission and response rates in week 12 ¹⁾



In week 12 after treatments, the clinical efficacy was evaluated by a partial Mayo score (DAI without endoscopy). Three of the 12 patients (25%) treated with Immunopure were in remission, as compared to 2 of 10 (20%) treated with prednisolone. In 9 (75%) and 5 (50%) of the patients DAI decreased by more than 1, respectively, classifying them as responders. Thus, the study has proven that the efficacy of the treatment with Immunopure was equivalent to prednisolone.



Adverse events

Immunopure There was no severe event in this study.

Prednisolone One symptomatic infection with *Clostridium difficile* occurred.

References:

1) Kruis, W; Nguyen, P; Morgenstern, J. Novel Leukocyte/Thrombocyte Apheresis for Induction of Steroid-Free Remission in Ulcerative Colitis: A Controlled Randomized Pilot Study. *Journal of Crohn's and Colitis*, 2019, 13(7), 949-953.



Specifications

| | | |
|-----------------------|-----------------|---------------|
| Adsorbent | Material | Polyarylate |
| Container | Material | Polycarbonate |
| | Length | 189 mm |
| | Diameter | 59 mm |
| Priming volume | | 135 mL |
| Sterilization | | Gamma rays |

Manufacturer

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