

Research Article

Study of the Bioavailability of Methyluracil *in Vitro* and Its Biocompatibility *in Vivo* in the Composition of Polyurethane Implant Material

Dmytro Kuliesh^{*} , Ludmila Nechaeva , Vira Hrytsenko , Rita Rozhnova 

Department of Medical Polymers, Institute of Macromolecular Chemistry of the National Academy of Sciences of Ukraine, Kyiv, Ukraine

Abstract

The development of polymer implant materials for use as drug carriers is an urgent task today. The aim of this work was to study the bioavailability of methyluracil when immobilized on a polyurethane carrier *in vitro*, as well as to study the biocompatibility of the obtained material when conducting model operations on experimental animals *in vivo*. As a result of the conducted studies, it was shown that the bioavailability of methyluracil immobilized on a polyurethane carrier is 78.5%, its prolonged release into the model environment is observed for 84 days. At the same time, more than 50% of the injected methyluracil was released by the 14th day of the study, which can significantly contribute to increasing the efficiency of regenerative processes at the implantation site, especially in the initial stages of the postoperative period. The conducted model operations on experimental animals made it possible to establish that the developed composite material with methyluracil is biocompatible and bioactive. Implantation of polymer samples with prolonged release of methyluracil contributed to the reduction of alteration and exudation phenomena in the implant placement area, activation of regeneration processes, and the formation of a mature and thin capsule around the implant already in the early stages of the study.

Keywords

Polyurethane, Methyluracil, Bioavailability, Biocompatibility, Implantation

1. Introduction

The development of polymer implant materials for use as drug carriers requires an interdisciplinary approach, including chemistry, biology, pharmacology and bioengineering [1]. It is known that polymer carriers allow to significantly improve the efficiency of drug delivery, ensuring local and controlled drug delivery directly to the desired area of the body, minimizing systemic side effects and toxicity, significantly increasing the bioavailability of the active substance [2-4],

which opens up new opportunities for personalized medicine and increasing the effectiveness of treatment. A number of requirements are imposed on polymer implant materials, which they must meet. Among the main ones are biocompatibility and safety, the absence of an immune response [5, 6]. In recent years, polyurethanes (PU), due to their chemical stability, high biocompatibility and low cytotoxicity, have been widely used in biomedical purposes [7-10]. PU are a

^{*}Corresponding author: d_kulesh@ukr.net (Dmytro Kuliesh)

Received: 17 July 2025; **Accepted:** 4 August 2025; **Published:** 20 August 2025



Copyright: © The Author(s), 2025. Published by Science Publishing Group. This is an **Open Access** article, distributed under the terms of the Creative Commons Attribution 4.0 License (<http://creativecommons.org/licenses/by/4.0/>), which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited.

class of polymers that can act as drug carriers, and depending on their composition, they can be given the necessary properties and specified parameters [11, 12].

One of the drugs that is promising for the purposes of immobilization on a PU carrier, in our opinion, is methyluracil, which belongs to the group of tissue regeneration stimulants. It is widely used in medicine to accelerate wound healing, tissue repair and improve metabolic processes [13]. Methyluracil has immunomodulatory and anti-inflammatory properties, is often used in medicine to stimulate cellular metabolism and protein synthesis, which contributes to the rapid healing of wounds and damaged tissues. As part of implant materials, methyluracil can be used to improve biocompatibility and accelerate tissue regeneration processes during implant use. Therefore, the current task is the development of polymeric materials with methyluracil as implants for use in various fields of medicine, in particular, in reconstructive surgery.

Considering the above, the purpose of this work was to study the bioavailability of methyluracil when immobilized on a PU carrier *in vitro*, as well as to study the biocompatibility of the obtained material when performing model operations on experimental animals *in vivo*.

2. Materials and Methods

2.1. Materials

The starting materials for obtaining composite materials were: polymer base - oligoetheretherethanediisocyanate, polymerization accelerator (PA-606/2) - 2,4,6-tris(dimethylaminomethyl)phenol. Methyluracil - a drug with regenerating, anti-inflammatory and immunostimulating properties - was used as a biologically active substance for immobilization on a polymer carrier. Polymer compositions with methyluracil in an amount of 2 wt. % were obtained, as well as PU samples without methyluracil as a control. Polymer compositions were obtained by sequential mechanical mixing of oligoetheretherethanediisocyanate, methyluracil, PA 606/2 and distilled water at room temperature. The resulting mixture was poured into fluoroplastic molds and dried in a thermostat at a temperature of 70°C. The cured composite materials had the appearance of fine-pored elastic sponges.

2.2. Drug Release of Methyluracil

It was of great interest to investigate the bioavailability of methyluracil immobilized on a PU carrier, using spectrophotometric determination of the dynamics of methyluracil release into a model environment. For this purpose, the obtained control polymer samples and polymer samples with methyluracil in an amount of 2 wt. % were placed in jars with ground stoppers, 40 ml of distilled water were added and incubated in a thermostat at a temperature of 37 ± 1 °C. The

obtained extracts were periodically drained and the optical density was recorded on a Specord M40 spectrophotometer in cuvettes with an optical layer thickness of 10 mm. During the study, distilled water was replaced with fresh water. An extract from a polymer sample without methyluracil was used as a control solution. UV absorption spectra of the studied methyluracil solutions before and after incubation of polymer samples under the specified conditions were identical to the spectrum of methyluracil itself.

The absorption spectrum of methyluracil had a maximum at a wavelength of $\lambda = 260 \pm 1$ nm. To verify the implementation of Beer's law and to construct a calibration graph of the dependence of the optical density of solutions on their concentration, a series of aqueous methyluracil solutions with concentrations: 0.00025; 0.0005; 0.00075; 0.001; 0.00125% was prepared. The calibration graph was a straight line in the entire range of studied concentrations.

The amount of methyluracil (MU) released from polymer samples was calculated by the formulas (1) and (2):

$$MU(mg) = C \cdot V \cdot n \quad (1)$$

$$MU(\%) = \frac{MU(mg)}{m} \cdot 100\% \quad (2)$$

where:

MU (mg) - the amount of methyluracil released, mg;

C - the concentration of methyluracil found from the calibration graph, %;

V - the volume of the solution in which the leaching took place, ml;

n - the degree of dilution of the solution during the analysis, times.

2.3. Animal Studies

2.3.1. Implantation Test

To study the interaction of the developed polymer materials with the tissues of a living organism, an implantation test was performed and cellular reactions in the tissues surrounding the implants of experimental animals were studied. The experiment was conducted on laboratory rats of the Wistar line weighing 180-210 g. All manipulations with experimental animals were carried out in compliance with the principles set forth in the European Convention for the Protection of Vertebrate Animals used for Experimental and Other Purposes [14] and in accordance with the Law of Ukraine "On the Protection of Animals from Cruelty" No. 3447-IV of February 21, 2006. Model operations were performed under aseptic conditions. After processing the surgical field, polymer samples without methyluracil and samples with 2 wt.% methyluracil measuring 5 x 10 mm were placed subcutaneously in the back area of experimental animals without additional fixation, to exclude the influence of the suture material

on the wound process. Animals were withdrawn from the experiment by ether overdose for 7, 14, 30 and 90 days.

2.3.2. Histological Evaluation

The experimental material (polymer sample with surrounding connective tissue) was fixed in 10% formalin solution and embedded in paraffin after histological processing according to the standard method [15]. Sections 10-15 μm thick were stained with hematoxylin and eosin. Analysis of cellular reactions and assessment of biocompatibility of polymer materials was carried out by examining histological preparations using light microscopy - microscopes "Mik-med-2", Carl Zeiss Primo Star, microphotography was carried out using a Canon PowerShot A640 camera with a Soligor Adapter Tube for Canon A610/A620 52 mm Tele.

3. Results and Discussion

3.1. Study of Drug Release of Methyluracil from Polyurethane Compositions in Vitro

As a result of the conducted studies, it was found that 78.5% of methyluracil immobilized on a PU carrier was released in a sustained manner over 84 days (Figure 1). At the same time, the "shock" dose (more than 50% of the administered methyluracil) was released by the 14th day of the study, which will probably allow to immediately achieve a high therapeutic concentration and lead to a rapid achievement of the therapeutic effect, increasing the efficiency of regenerative processes at the implantation site in the initial stages of the postoperative period.

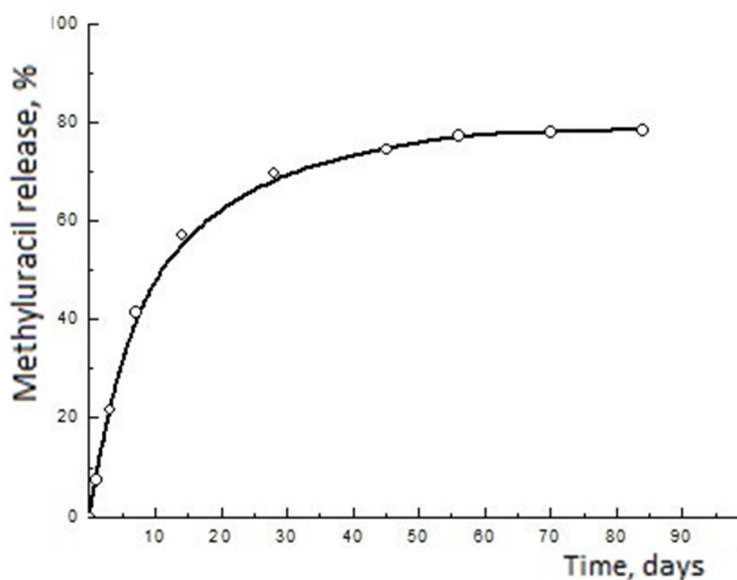


Figure 1. Drug release of methyluracil from PU carrier.

3.2. Implantation Test

During the histological study, the behavioral reaction of animals, their external condition, and the postoperative field were studied. Daily visual assessment of the epithelial reaction at the surgical site showed that the wound healed 3-5 days after surgery without signs of an inflammatory reaction. According to morphological signs, practically no degenerative changes, tumors, or tissue necrosis were detected either in the short-term or in the long-term postoperative period. Throughout the experiment, the implanted polymer samples were palpated through the skin of the animals. The implantation of the studied samples did not cause aggression or changes in the behavior of the experimental animals. The main attention in histological studies was paid to signs of the development of inflammatory phenomena in the area of implantation of polymer samples at the "implant - tissue" inter-

face. Macroscopically, connective tissue was detected around the implanted samples at all times of the study, which separated the implanted samples from the surrounding tissues and did not differ in color and structure from tissues further from the implantation site.

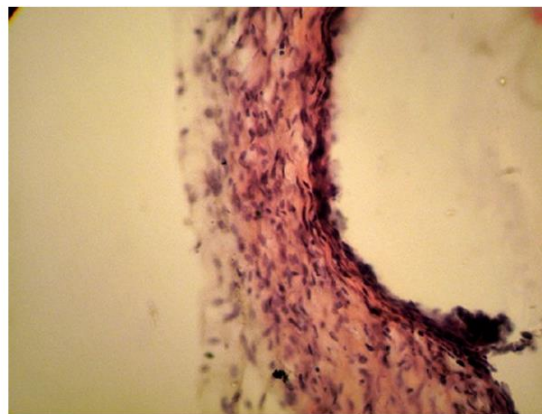
Microscopically, 7 days after surgery, a connective tissue capsule (CTC) was observed around the PU sample without methyluracil, in the thickness of which many blood vessels, poorly differentiated cellular elements, young forms of fibroblasts, and immature collagen fibers were observed (Figure 2a). Lymphocytic and neutrophilic infiltration was noted, activation of monocyte-macrophage elements was observed, which were present in large numbers. 7 days after surgery, in rats implanted with PU samples with methyluracil, the CTC was more mature, thinner, and less dense than around the control PU samples. The main cellular elements were spindle-shaped fibroblasts oriented along the implanted material (Figure 2b). In places, the capsule was less mature, young

forms of fibroblastic elements and slight neutrophilic infiltration were present. Monocyte-macrophage elements were present in small quantities and in separate areas of the capsule. Blood vessels were characterized by normal trophism, but

there were several vessels with complications of microcirculatory processes, which was expressed in the marginal location of formed blood elements and the expanded lumen of the vessels themselves.



a) pronounced round cell reaction around the control sample. $\times 160$

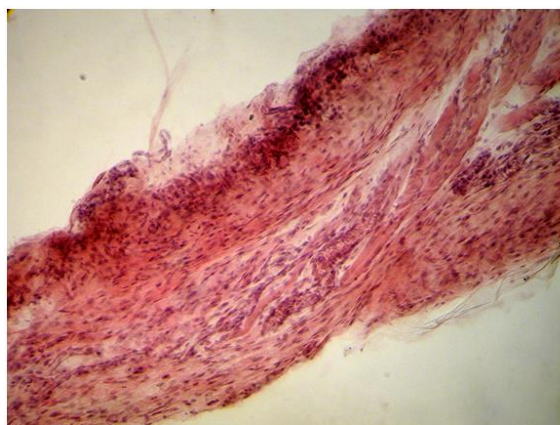


b) more mature and formed capsule around the sample of PU with methyluracil. $\times 400$

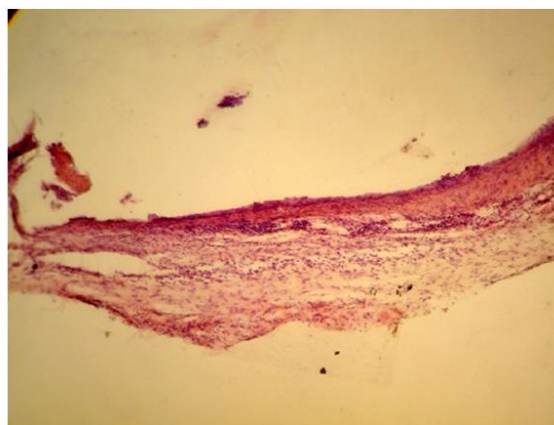
Figure 2. Capsules around the implanted samples on the 7th day after surgery. Hematoxylin-eosin staining.

14 days after surgery in animals implanted with PU samples without methyluracil, the CTC was quite thick, not mature enough with a large number of blood vessels (**Figure 3a**). Young forms of fibroblastic elements were observed, lymphocytic-macrophageal and partially neutrophilic infiltration was preserved. All this indicated that the intensity of the tissue reaction to the implantation of polymer samples remained quite high. 14 days after surgery around the PU samples with methyluracil, the CTC was significantly thinner than around the PU samples (control) at a similar time point of the study. It consisted of two layers that were radically different from each

other. The first (inner) layer consisted of dense unformed connective tissue, which was characterized by a quantitative predominance of fibers over the ground substance and cellular elements. The second (outer) layer had in its structure mainly round-cell elements, among which monocytic-macrophage elements prevailed (**Figure 3b**). Macrophages migrated to the source of irritation - a foreign body, phagocytizing the products of tissue decay, cellular detritus. Nearby was a small number of blood vessels, their microcirculatory processes were within normal limits.



a) capsule with a large number of blood vessels. $\times 400$



b) capsule around the sample of PU with methyluracil. $\times 160$

Figure 3. Connective tissue capsule around implanted samples on day 14 of the experiment. Hematoxylin and eosin staining.

30 days after surgery, a rather thin, sometimes immature CTC was observed around the PU samples without methyluracil, which was represented by young forms of fibroblastic elements, there was local leukocyte infiltration, cellular detritus (Figure 4). In other areas, the capsule was represented by bundles of mature collagen fibers and spindle-shaped fibroblasts between them, oriented along the polymer implanted material. Blood vessels were present in large numbers, most of them were full-blooded and dilated. 30 days after surgery, a mature, slightly thickened CTC was observed around the PU sample with methyluracil, which was characterized by a high degree of maturity, consisted of bundles of collagen fibers and mature spindle-shaped fibroblasts between them. Infiltration by neutrophilic and lymphocytic elements was absent. The monocyte-macrophage reaction was moderate, compared to the previous period of the study. Exudative processes around the placed implant were not observed, but an active process of vascularization of young blood vessels with normal trophism was observed.

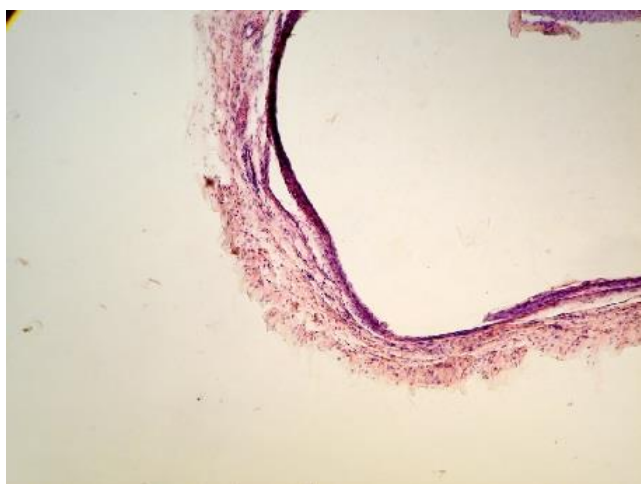


Figure 4. Connective tissue capsule around the PU sample (control) 30 days after surgery. Hematoxylin and eosin staining. $\times 160$.

90 days after surgery, the capsule density around the control PU samples decreased due to remodeling (remodulation) and involution of the connective tissue with thinning of the capsule, which was characterized by a high degree of maturity. Leukocyte and lymphocyte infiltration were absent. A dense network of blood vessels was observed in the adjacent connective tissue. All of them were characterized by a thickened wall of elastic fibers, were without complications of microcirculation, i.e. not filled with blood, without elements of stasis and thrombosis. 90 days after surgery, a similar histological picture was observed around the PU sample with methyluracil as around the PU sample without methyluracil. Mature CTC was observed, consisting of bundles of collagen fibers oriented along the polymer samples and spindle-shaped fibroblasts. The capsule was without signs of leukocyte and

lymphocyte infiltration, the monocyte-macrophage reaction was insignificant. A dense network of blood vessels was also observed in the adjacent connective tissue, without complications of microcirculation. Macrophage infiltration around all samples was noticeable, which is apparently associated with the processes of polymer biodegradation. Thus, according to the results of the histological studies, it was established that around the samples of PU with methyluracil, already on the 7th day after the operation, the CTC was more mature and much thinner, while the process of capsule maturation and reduction of the intensity of the inflammatory reaction in the control group progressed only to the 30th day after implantation. When all experimental samples were implanted into the body of experimental animals, inflammatory phenomena associated with surgical trauma and the presence of the body itself were observed in the early stages. Such reactions were natural in the early stages of the study and were consistently replaced by a decrease in the area of infiltrates, timely reduction of the vascular bed, which ended with the formation of mature CTCs around the implants without the phenomena of chronic inflammation. Around the samples of PU with methyluracil 30 and 90 days after the operation, a significant increase in not only the fibrous component, but also active angiogenesis was noted, which indicated active proliferative processes in the implantation zone. Against the background of CTC maturation in the control group, there was a slight increase in the intensity of the inflammatory reaction due to the appearance of foci of leukocyte and lymphocyte infiltration.

4. Conclusion

As a result of the work, was established that the bioavailability of methyluracil immobilized on a polyurethane carrier is 78.5%, and its prolonged release into the model environment is observed for 84 days. At the same time, more than 50% of the injected methyluracil was released by the 14th day of the study, which can significantly contribute to increasing the efficiency of regenerative processes at the implantation site, especially in the initial stages of the postoperative period. Model operations on experimental animals made it possible to establish that the developed composite material with methyluracil is biocompatible and bioactive. Implantation of polymer samples with prolonged release of methyluracil contributed to the reduction of alteration and exudation phenomena in the implant placement area, activation of regeneration processes and the formation of a mature and thin capsule around the implant already in the early stages of the study.

Abbreviations

PA-606/2	Polymerization Accelerator (2,4,6-tris(dimethylaminomethyl)phenol)
CTC	Connective Tissue Capsule

PU Polyurethane
 MU Methylluracil

Conflicts of Interest

The authors declare no conflicts of interest.

References

- [1] Stewart SA, Dom ínguez-Robles J, Donnelly RF, Larrañeta E. Implantable Polymeric Drug Delivery Devices: Classification, Manufacture, Materials, and Clinical Applications. *Polymers*. 2018; 10(12), 1379. <https://doi.org/10.3390/polym10121379>
- [2] Pratap Singh L., Patro L. R., Sayeed M., Rani mandadi S., Pant N. C., Panda C., Aparna T. N., Vodeti R., Das C. Polymeric Drug Delivery Systems: Chemical Design, Functionalization and Biomedical Applications. *J. Chem. Rev.* 2025, 7(3), 421-451. <https://doi.org/10.48309/jcr.2025.516027.1435>
- [3] Sharma P, Jain V, Tailang M. Selection and Role of Polymers for Designing of a Drug Carrier [Internet]. *Drug Carriers*. IntechOpen; 2022. Available from: <http://dx.doi.org/10.5772/intechopen.103125>
- [4] Sung, Y. K., Kim, S. W. Recent advances in polymeric drug delivery systems. *Biomater Research*. 2020, 24, 12. <https://doi.org/10.1186/s40824-020-00190-7>
- [5] Galatenko N. A., Rozhnova R. A. Biologically active polymeric materials for medicine, Kyiv, Naukova Dumka. 2013, 211 p.
- [6] Gryn S. V., Synyugina A. T. *Polymers for medical purposes*, Kyiv. 2023, 88 p.
- [7] Wienen D., Gries T., Cooper S. L., Heath D. E. An overview of polyurethane biomaterials and their use in drug delivery. *Journal of Controlled Release*. 2023, 363, 376-388. <https://doi.org/10.1016/j.jconrel.2023.09.036>
- [8] Cui M., Chai Z., Lu Y., Zhu J., Chen J. Developments of polyurethane in biomedical applications: A review, *Resources Chemicals and Materials*. 2023, 2(4), 262-276. <https://doi.org/10.1016/j.recmm.2023.07.004>
- [9] Sobczak, M.; Kędra, K. Biomedical polyurethanes for anti-cancer drug delivery systems: a brief, comprehensive review. *Int. J. Mol. Sci.* 2022, 23, 8181. <https://doi.org/10.3390/ijms23158181>
- [10] Drożdż K., Gołda-Cępa M., Brzychczy-Włoch M. Polyurethanes as Biomaterials in Medicine: Advanced Applications, Infection Challenges, and Innovative Surface Modification Methods. *Advancements of Microbiology*. 2025, 4(63), 223-238. <https://doi.org/10.2478/am-2024-0018>
- [11] Wang W., Wang C. Polyurethane for biomedical applications: A review of recent developments. In the design and manufacture of medical devices. 2012. 115-151. <https://doi.org/10.1016/B978-1-907568-72-5.50003-0>
- [12] Bose N., Rajappan K. Summary on polyurethane-based drug delivery system in perspective for future implantable drug system. *International Journal of Polymeric Materials and Polymeric Biomaterials*. 2024, 73(18), 1629-1648. <https://doi.org/10.1080/00914037.2023.2301647>
- [13] Svitlana V. Shishkina, Anna M. Shaposhnik and Victoriya V. Dyakonenko et al. New Polymorphic Modifications of 6-Methyluracil: An Experimental and Quantum Chemical Study. *ACS Omega*. 2023. Vol. 8(23): 20661-20674. <https://doi.org/10.1021/acsomega.3c01231>
- [14] European convention for the protection of vertebrate animals used for experimental and other scientific purposes. Council of Europe, Strasbourg. 1986, 53 p.
- [15] Bahrii M. M., Dibrova V. A., Popadynets O. H., Hryshchuk M. I. *Metodyky morfolohichnykh doslidzhen*. Vinnytsia: Nova Knyha. 2016, 328 p.