


# Remodeling of the Fibula Stump After Transtibial Amputation

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**Aim:** To study the peculiarities of peroneal stump remodelling after transtibial amputation in the process of prosthesis usage.

**Material and Methods:** A histological study of the ends of the stumps of the fibula in 68 patients was performed. Terms after amputation: 2–8 years.

**Results:** In the 1st group the stumps with the reparative process completion were formed. In the 2nd group there were sharp disturbances of the reparative process with the formation of the cone-shaped end. In the 3rd group there was a pronounced periosteal bone formation with changes in the shape and structure of bone tissue and incompleteness of the reparative process.

**Conclusion:** Absence of balloting of the fibula stump and dense overlapping of the medullary cavity by muscles promotes complete remodelling of the fibula remnant with preservation of its organicity. Pathological remodelling of the fibula stump occurs due to its hypermobility, repeated traumatization of the forming regenerate, neuritis of the peroneal nerve, osteogenesis disorders and structural and functional mismatch of the bone tissue to the loading conditions in the prosthesis. Morphological signs of pathological remodelling are the lack of completion of reparative regeneration, intensive bone tissue remodelling lasting for years with pronounced resorption and appearance of immature bone structures, fractures of the cortical diaphyseal layer, residual limb deformities with formation of a functional regenerates, narrowing and closure of the medullary canal with conglomerate with soft tissue inclusions. The anatomical inferiority of bone tissue formed in the process of remodelling of the fibula remnant creates a threat of stress fracture.

**Keywords:** balloting, fibula stump, pathological remodeling, transtibial amputation

## Introduction

The methods, techniques and results of transtibial amputations are well described in the available literature,<sup>1–9</sup> as well as the effect of local, due to neuroma, and phantom pain syndrome on the quality of life of patients.<sup>8,9</sup> Emphasis is placed on the excessive mobility of the residual fibula, its valgus deviation, pain syndrome on the outer surface of the residual limb, and difficulties or impossibility of prosthesis. These works did not reflect the issues of anatomical state of the remnant of this bone and reparative and restructuring processes occurring after amputation. This problem is partially covered in experimental studies.<sup>10,11</sup> However, the data given by the authors concern the formation of the femoral stump in animals and do not touch upon the issues of changes in the structural organisation of bone tissue under load in the prosthesis. Recently, the issues of optical identification of model parameters,<sup>12</sup> fabrication of pneumatic reconfigured sockets for proper stump fit,<sup>13</sup> and characterization of soft tissue features after tibial amputation<sup>14</sup> have been developed, which may in the future affect both bone structure and prosthetic outcomes. The available studies of the peroneal bone tissue after transtibial amputation only state the disturbance of bone formation in the form of acuminosity or club-like shape and phenomena of peroneal nerve neuritis.<sup>15</sup> By increasing the total cross-sectional area of the residual limb bones together with the tibia, the fibula becomes even more vulnerable than the tibia. This is facilitated by frequent damage to the interosseous membrane, the anterior and posterior intermuscular septa, which limit the mobility of the fibula, and muscle atrophy during amputation.<sup>15</sup> Excessive mobility of the fibula stump results in trauma to the tissues, irritation of the peroneal nerve, and complication of prosthetics.<sup>10</sup> As in fractures,<sup>16</sup> in conditions of residual limb mobility, repeated

traumatisation of the residual limb can hinder osteogenesis both by mechanical impact on the regenerate forming and by damage to the microcirculatory pathways. According to the data<sup>17</sup> 81% of patients with amputations of the tibia as a result of pain and limited prosthesis use require revision interventions at the end of the fibula.

Aim of the work: to study the peculiarities of peroneal stump remodelling after transtibial amputation in the process of prosthesis use.

## Materials and Methods

Under our supervision were 68 male patients aged 19–40 years with transtibial amputations who underwent revision surgery for various reasons related to the impossibility or significant limitations of using the prosthesis. Cause of primary amputation: combat injuries (gunshot, mine blast, multiple rocket launcher systems, compression syndrome) – in 58, peacetime injuries – in 8, thermal injuries – in 2. The amputation was performed up to 2 years ago – 12 people, up to 5 years ago – 49 people, and over 5 years ago – 7 people. Prior to admission to the clinic, all 68 people were wearing a deep-fitting prosthesis.

Patients were selected by random sampling based on the need for surgical treatment.

Inclusion criteria: at least one year of prosthesis use; high muscle position; soft tissue excess; massive scars fused to the bone; valgus deviation of the fibula stump; hypermobility of the fibula stump; improper resection; painful neuroma. As a rule, a combination of several malformations was observed.

Exclusion criteria: purulent and inflammatory skin diseases, chronic osteomyelitis of the bone stump, non-prosthetic stump, BMI <20 kg/m<sup>2</sup>, diabetes mellitus, glucocorticosteroids, bisphosphonates within the last 3 months, chronic kidney disease, hyperthyroidism and hypothyroidism, chronic heart failure, oncological pathology, tuberculosis, systemic diseases.

The main complaints were pain, instability and limited time of use of the prosthesis. In 20 people (29.4%), the pain was osteomedullary in the fibula remnant with a feeling of its bulging or numbness. In 18 people (26.5%), it was combined with pain on the outer surface of the stump with numbness and paresthesias. The association of pain with prosthesis use was noted by all patients with conical and club-shaped fibula stumps.

Clinically, 38 patients (55.9%) had excessive mobility in the frontal and sagittal planes within 2–3.8 cm with light efforts applied to the end of the stump. Persistent valgus deviation of the stump was observed in 18 patients (26.5%). There was also uneven thickening of the biceps tendon, a bursa at the end of the fibula stump, ulcers and scars on the skin. The dense fibrous layer at the end of the stump was represented by thinned skin.

Amputation stumps at the level of the upper third of the tibia were present in 24 patients (35.3%), and at the border of the upper and middle third – in 44 (64.7%). Bilateral amputations occurred in 8 patients (11.7%).

Radiographically, 30 patients (44.1%) had stumps of normal shape (group 1), typical for this level. In 18 patients (26.5%), the end of the stump was conical in shape (group 2), and in 20 patients (29.4%), the end of the stump was club-shaped (group 3).

In patients of groups II and III, during revision interventions, in addition to surgeries on the stump bones, the valgus deviation and excessive mobility of the fibula were eliminated by stabilizing the long fibula muscle attached to the end of the fibula and the outer surface of the tibia (myodesis). The anterior and posterior intermuscular membranes were restored. Patients of group I underwent revision surgery for tibial stump defects with its shortening and corresponding alignment of the fibula length. The fibula stump was stabilized in the same way as in groups II–III. The final myoplasty was performed in the same way for all three groups. A transverse channel 0.6–1 cm long was made in the upper part of the sawn-off tibial crest without penetrating the marrow canal. A long resorbable thread was passed through the channel. One of its free ends was sutured to the anterior edge of the gastrocnemius muscle, the other to the edge of the medial head of the gastrocnemius muscle. The ligature was tied, covering the ridge. The calf muscle was placed over these muscles and bones, fixing it percutaneously to the holes in the tibia. An elastic muscle stump was formed on the operating table.

The results of surgical treatment were evaluated in 45 patients in the preoperative and postoperative (12 months) periods by X-ray and by the average time of use of the prosthesis during the day (hours). Patients described the duration of prosthesis use in a questionnaire.

The results of the histological study are based on the material taken during revision interventions on the residual limb. The material included the ends of peroneal and tibial bone filaments, their soft tissue border, and sections of the resected peroneal nerve. The preparations were fixed in 12% neutral formalin solution and decalcified in 8% nitric acid solution. After degreasing and dehydration in acetones and alcohols of increasing strength and alcohol-ether, a sagittal cut through the middle of the bone was made, which was blocked in celloidin. Sections 15–30  $\mu\text{m}$  thick were stained with haematoxylin and eosin and Van-Gizon picrofuchsin. When studying the preparations, attention was paid to the changes in the cortical diaphyseal layer, the shape of the end section, the character of bone structures composing it, the presence of a closed medullary cavity, the formation of the closing bone plate, the condition of tissues inside the medullary canal, and the character of the soft tissue covering of the stump end. Nerve fibre sections were stained with hematoxylin-eosin and impregnated with silver.

The morphometric study was performed using digitized images of the preparations on a Pannoramic scanner (3DHISTECH, Hungary) using the image analysis programs Orbit Image Analysis and Pannoramic Viewer. To evaluate the amount and structure of bone tissue, the area (S) of osteogenic, chondrocytic, and fibrous tissue in the specimen and their percentage of the specimen area were determined.

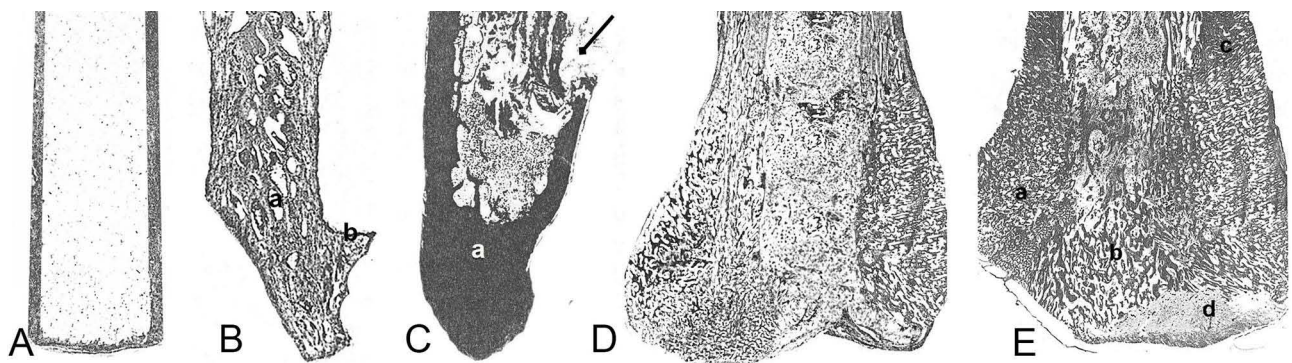
## Statistical Analysis

The statistical processing of the study results was performed using the software package “MS Excel XP” and “IBM SPSS Statistics” 28.0.1.0. The frequency of quantitative indicators is presented in absolute (n) and relative (%) frequencies. The normality of the distribution of the obtained results was determined, which are presented as mean (M)  $\pm$  standard deviation (SD). The study of the significance of differences between the means by comparing the variances was performed using One Way ANOVA. The division of the total variance into several sources made it possible to compare the variance caused by the difference between the groups with the variance caused by within-group variability. A difference of  $p < 0.05$  was considered statistically significant. Pairwise analysis was performed using the Student’s parametric *t*-test for related samples.<sup>18,19</sup>

## Results

### Group I, Stumps of Regular Anatomical Shape (30 Observations)

In all observations, the stump had a flat base with a closure plate of compact bone tissue at the end (Figure 1). Moderately pronounced reparative processes were observed in the cortical diaphyseal layer with insignificant bone resorption mainly along the course of the vascular channels. The contours of the tubular bone and the structure of compact bone in the cortical layer were preserved. No fresh bone-forming processes were observed on the endosteal and periosteal surfaces of the residual limb. Bone marrow tissue was represented by fatty bone marrow. The dense fibrous



**Figure 1** Histotopograms of peroneal stump ends (haematoxylin and eosin staining,  $\times 2.5$ ): (A) Cylindrical shape with formation of bone closure plate from mature bone tissue; (B) Cone-shaped with congestion of the medullary canal (a) and a small periosteal regenerate (b); (C) Cone-shaped with occlusion of the medullary canal (a), violation of the integrity of the cortical diaphyseal layer (arrow); (D) Club-shaped with full length occlusion of the medullary canal with dense fibrous tissue (a), endosteal-periosteal regenerate at the end (b); (E) Club-shaped: resorption of the cortical diaphyseal plate and its replacement by endosteal-periosteal regenerate (a); endosteal bone beams (b); spongy cortical diaphyseal plate in the proximal region (c); fibrous cartilaginous tissue edging the end of the stump (d).

layer at the end of the residual limb had an organ structure of thick bundles of collagen fibres with an orderly distribution of vessels between them. The peroneal nerve at the end was thickened at the end, dystrophically changed, without inflammatory phenomena.

## Group 2, Cone-Shaped Stumps (18 Observations)

In addition to conicality, there was a curvature of the truncated bone axis with asymmetry and shortening of the formed residual limb due to significant bone resorption at the end of the file (Figure 1). In part of the observations, immature bone structures with fibrous cartilage inclusion were detected on the lower surface of the cone-shaped end, and in the other part, the endosteal regeneration from immature bone or fibrous bone tissue occurred over a large length. The formation of a bony closure plate was not observed in any of the cases. The cortical diaphyseal layer was thickened, thinning in some places, sharply rarified, spongy, and underwent active focal osteoclastic resorption. In 6 observations, fresh and healing fractures with the appearance of endosteal and periosteal formed immature bone beams were seen near the end (Figure 1). Necrobiotic changes and a sharp increase in osteoclastic resorption were revealed. In the areas of thinning of the cortical diaphyseal layer, cellular fibrous osteoblastic tissue with the presence of osteoclasts was detected. In other sections, pronounced dystrophic changes and foci of necrosis were noted (Figure 2). In the area of resorption there were diffusely located macrophages, lymphoid and plasma cells. On the endosteal and periosteal surfaces there were fresh bone tissue overlays, foci of immature bone tissue, atrophy. In the narrowed medullary canal there was edematous loose fibrous tissue with areas of fibroreticular and yellow edematous bone marrow, rare immature bone beams. The fibrocartilaginous covering adjacent to the end included areas of vessel-rich loose fibrous tissue with disordered course of thin collagen fibres, focal accumulations of leukocytic cellular elements, whole conglomerates of vessels. When examining the peroneal nerve, pronounced oedema of epineurium, perineurium, endoneurium, infiltration of the trunk with lymphocytes were observed.

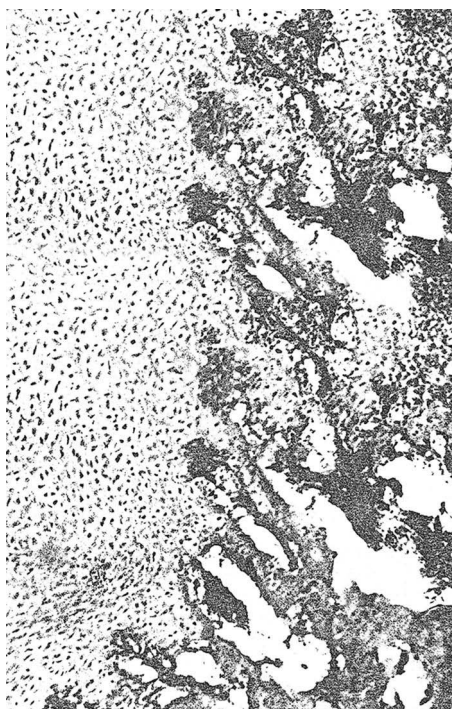
## Group 3, Club-Shaped Stumps (20 Observations)

The club-like appearance of the residual limb was due to significant resorption of the cortical diaphyseal layer and fusion of periosteal and endosteal regenerates (Figure 1D and E). The cancellous bone tissue of the regenerates is immature, with phenomena of intensive remodelling. In some observations they are covered with hyaline cartilage, in others – with fibrous cartilage (Figure 3). Loose fibrous and fibrous tissue was found in the interbar spaces of the bone-cartilaginous regenerate (Figure 4). Dystrophic changes of the cartilage edges were revealed. Above the regenerates, the cortical diaphyseal layer was spongy. There were areas of immature bone beams, large foci of osteoclastic resorption and transformation (Figure 5). The transformed part underwent intensive remodelling, wide medullary cavities were formed,

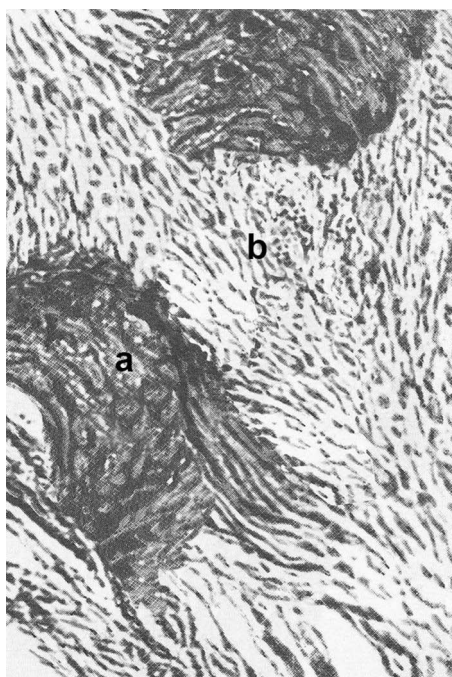


**Figure 2** Microphotograph. Dystrophic changes and necrosis of bone tissue. Haematoxylin and eosin staining. x120.



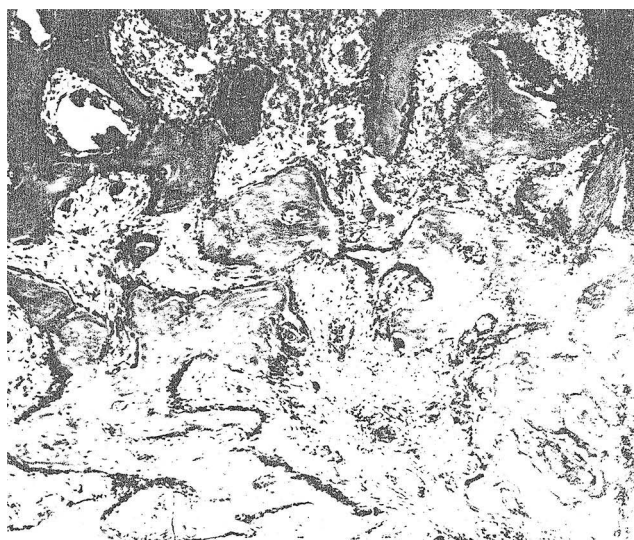


**Figure 3** Microphotograph. Spongy tissue of periosteal regenerate bordered by hyaline cartilage. Haematoxylin and eosin staining. x90.

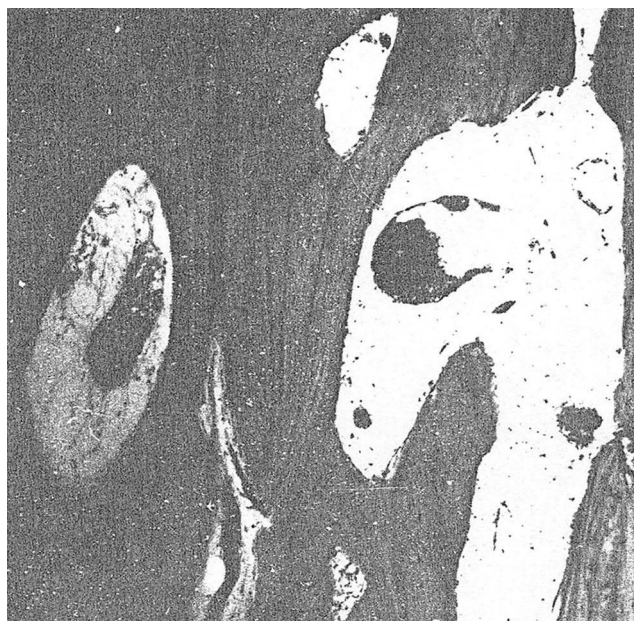


**Figure 4** Microphotograph. Dissolution of bone beams of cancellous bone (a) bone-cartilaginous regenerate and its replacement by fibrous tissue (b). Haematoxylin and eosin staining. x130.

changing the structure of the compact bone (Figure 6). In some cases, individual immature bar structures of the forming bone closure plate were detected along a small length of the residual limb base; in other cases, the endosteal regenerate had a spongy structure represented by immature bone beams, osteogenic tissue, and cartilage interlayers. Fresh periosteal and endosteal bone formation was determined, represented by a dense network of bone beams, amenable to remodelling.



**Figure 5** Microphotograph. An area of active osteoclastic resorption of compact bone. Haematoxylin and eosin staining. x120.



**Figure 6** Microphotograph. Resorption of compact bone tissue. Full blood vessels. Haematoxylin and eosin staining. x80.

The muscles surrounding the stump are severely swollen. The soft tissue vessels are deformed. Their walls are thickened. Fibrous tissue, structures such as villous formations of the joint bag, and disintegrating blood cells were detected in the preparations of the fibrous rim of the end surface of the stump. The peroneal nerve was thickened at the end. No inflammatory changes were detected.

In the morphometric study of tissue structures in patients of group I, the content of osteogenic tissue was significantly higher compared to patients of groups II and III ( $p < 0.05$ ), indicating a certain completeness of bone formation processes in the tissues of the amputation stump (Table 1). Intensive bone remodelling and incomplete reparative processes at the end of the fibula stump were characterised by a decrease in the content of osteogenic and a significant increase in fibrogenic structures in the histological material of patients of group II ( $p_{I,II} < 0.05$ ;  $p_{II,III} < 0.05$ ) and chondrogenic - patients of group III ( $p_{I,III} < 0.01$ ).

**Table I** Quantitative Evaluation of the Tissue Structures of the Amputation Stump of the Fibula

Observations	Tissue Structures, %		
	Osteogenic, M±SD	Chondrogenic, M±SD	Fibrogenic, M±SD
I group (n=30)	89.13±5.24	3.07±2.68	7.8±3.65
II group (n=18)	73.33±5.58	11.92±3.41	17.49±3.04
III group (n=20)	73.95±4.63	16.15±3.91	8.71±3.04
Significance degree (p-value)	p<0.001	p<0.001	p<0.001
F - Fisher's criterion, actual	75.3011	83.8624	19.2367
F - Fisher's criterion, critical	3.1381		
	Pairwise comparison		
P <sub>I,II</sub>	<0.05	<0.05	<0.05
P <sub>I,III</sub>	<0.05	<0.01	>0.05
P <sub>II,III</sub>	>0.05	>0.05	<0.05

## Discussion

Bone stump remodelling is a complex repair process in which, as in fractures, it is necessary to distinguish between a reparative reaction and a remodelling process.<sup>16</sup> The reparative reaction gradually progresses to the remodelling process, which ends only with the completion of bone formation as an organ. The remodelling cycles continue until the restoration of stable relations between the blood supply pools of bone, which ensure the preservation of tissue homeostasis and bone structure.<sup>11,20</sup>

For normal healing of the end of the bone stump, it is necessary to achieve such conditions as tight closure of the medullary canal with muscle or graft and rest during the period of active regeneration.<sup>10</sup> Unfortunately, achieving immobility of the fibula end is a difficult task. In almost all cases of cone-shaped and club-shaped stumps, we observed partial or complete damage to the interosseous membrane and the anterior and posterior intermuscular septa.

A cylindrical stump with a preserved structure of the cortical diaphyseal layer, a medullary canal traceable throughout, and rapid healing of the latter along the edge of the fillet with a closure bone plate should be considered an organotypic stump of the diaphyseal section of a tubular bone. A characteristic feature of such a residual limb is the completion of intensive reparative processes.<sup>20</sup>

In group 1, when the muscle plasty was correctly performed, limiting the balloting of the fibula remnant and overlapping its medullary canal, the residual limb was a painless organ with the completion of reparative processes, stable shape, a bony closure plate at the end, normal medullary content, moderately ongoing remodelling processes and a dense fibrous coating at the end of the bone, allowing painless loading in a full-contact prosthesis. The absence of fresh endosteal and periosteal bone formation and active remodelling at a distant time indicated the equilibrium of osteo-epariation and resorption processes and the completion of the reparative process. Thus, in this group, despite amputation and prosthetics that radically changed the anatomy and natural loading conditions, physiological remodelling led to the establishment of a new level of anatomo-functional compliance of the fibula stump.

The formation of the cone-shaped residual limb was influenced by loading and traumatization in the prosthesis with resorption of the end of the cortical diaphyseal layer and the resulting bone formation in response to the incomplete replacement of the resorbed bone. Atrophy of the cortical diaphyseal layer and endosteal regenerate developed against the background of attenuation of reparative processes and prevalence of bone and cartilage tissue resorption. These



processes were aggravated by pronounced phenomena of peroneal nerve neuritis. The resulting changes in bone structure had a persistent decompensated character.

The formation of a club-shaped residual limb with the formation of a voluminous osteochondral and fibrous callus at the initial stage was due to decompensation of the disturbed blood supply. Later, under the influence of prosthetics, the process progressed due to overstrain and activation of the already compromised bone remodelling. These data are in agreement with the opinion of,<sup>16</sup> who revealed that in an anatomically incomplete regenerate under the influence of overstrain a reparative reaction may occur, causing its rapid increase. It is caused by the impossibility of osteogenesis due to the destruction of sprouting vessels by the mobile end of the residual limb. The resulting anatomical inferiority due to repeated cycles of remodelling diminishes slowly and persists for years.

With continued functional load, overload, and irrational prosthetics, pathological bone tissue remodelling developed in groups 2 and 3, as evidenced by bone stump shape abnormalities, identified fresh endosteal and periosteal bone formation, active remodelling, fresh fractures, and healed fractures. Stress fractures occurred against the background of pronounced rarification of compact tissue. The presence of diffusely scattered macrophages and lymphoid-cell clusters among the osteoblastic tissue and in the superficial layers of the fibrous part of the regenerate is associated with reactive productive inflammation in response to excessive mobility and traumatisation of the residual limb, which to some extent stimulated the bone formation process. The presence of immature bone beams of intensive osteogenesis in the regenerate at the end of the residual limb also testifies to this fact.

The muscle tissue under the bone stump forms a solid dense fibrous tissue layer with an organ structure, which we observed at the end of the bone stump in all the observations of group 1. In groups 2 and 3 there was no such organicity. In 12 preparations (17.6%), this layer was easily wounded when using the prosthesis, often ulcerated, and probably influenced the deformation of the bone end on the basis of inflammation, which was confirmed by observations.<sup>21</sup>

The bone and underlying tissue regenerate formed during ballottisation of the fibula stump is non-organotypic. Its structure and shape can only conditionally correspond to the bone stump. Due to pronounced dystrophic and reconstructive processes lasting for years, accompanied by painful disorders, they worsen the quality of life and carry a threat of stress and traumatic fractures.

The conducted studies have shown the dependence of peroneal stump remodelling on its mobility, involvement of the peroneal nerve and prosthetics. Apparently, before prosthetics it occurred due to the arisen mismatch of microcirculation to the metabolic needs of bone tissue, and in the process of loading and traumatisation an anatomico-functional mismatch appeared in the prosthesis, accompanied by uneven distribution of mechanical stresses and appearance in the bone of areas of reduced and increased stress concentration and, accordingly, less and more intensive microcirculation.

The functional suitability of the residual limb includes its ability to enter into a “correct” relationship with the prosthetic socket.<sup>21,22</sup> It must have partial end-support and be able to withstand the uniform pressure exerted in the prosthetic socket. Group 1 residual limbs fulfilled these criteria. Without fulfilment of these conditions, walking was limited due to pain.

The clinical and morphological observations demonstrate the need for stabilisation of the fibula stump, tight overlap of the medullary cavity with sutured muscles during amputation, and careful fitting of the prosthesis.

Limitations of our study include the small sample size and short-term follow-up. A larger sample with a different start may reflect other possible problems.

In the long term after the surgical interventions, an increase in the average time of prosthesis use (ATPU) was noted in patients of all study groups (Table 2). In patients of group I, this indicator increased slightly, only by 9.23%, was equal to  $11.36 \pm 0.89$  hours and did not differ significantly ( $p=0.0568$ ) from the results obtained before the operation. Significant dynamics of growth of indicators was observed in patients of groups II and III, 77.17% and 50.77%, respectively. At the same time, there was a significant difference between the results of the pre- and postoperative periods ( $p<0.001$ ).

A slight percentage increase in the average time of prosthesis use in patients of group I can be explained by the features and nature of revision interventions. In contrast to patients of groups II and III, where pathological remodeling was in the first place, in patients of group I, revision interventions were performed for pathology of the tibial stump with its shortening and corresponding alignment of the fibula length.



**Table 2** Indicators of the Average Time of Prosthesis Use in Patients of the Study Groups Before and After Surgery

Indicators	I Group (n=15)		II Group (n=15)		III Group (n=15)	
	Before Surgery	After Surgery	Before Surgery	After Surgery	Before Surgery	After Surgery
ATPU during the day, hours (M±SD)	10.4±2.61	11.36±0.89	6.09±1.38	10.79±1.70	7.17±1.52	10.81±0.82
Abnormalities of ATPU during the day after surgery to the preoperative period, hours	0.96		4.7		3.64	
Improvement dynamics,%	9.23		77.17		50.77	
Variation coefficient (Vσ), %	15.0	8.0	18.67	7.12	16.59	8.11
Significance degree (p-value)	P=0.0568		p<0.001		p<0.001	
F - Fisher's criterion, actual	3.9468		107.3455		84.6259	
F - Fisher's criterion, critical	4.1959					

## Conclusions

1. Favorable course of peroneal stump remodeling is noted only under optimal conditions, which include preservation of the interosseous membrane, high truncation of the peroneal nerve, complete myoplasty with dense closure of the medullary cavity, which contributes to the restoration of microcirculation, timely and high-quality prosthetics with gradually increasing functional load.
2. Ignoring these conditions leads to hypermobility of the fibula stump, which causes local inflammatory complications, impaired regional blood circulation, damage to vascular connections between the bone and adjacent tissues, prolonged reparative osteogenesis, and repeated reparative cycles. Disturbances of microcirculation activate bone remodeling, which lasts for years and is aggravated by poor-quality prosthetics.
3. The combination of unfavorable pathogenetic factors leads to significant variability in the forms of truncated stumps (club-shaped due to pronounced periosteal bone formation, cone-shaped as a result of resorption and atrophy of the end of the bone stump, curvature of the axis, absence of bone closure plate, bone resorption, uneven reparative regeneration), as well as disturbance of tissue homeostasis and bone structure.
4. Stress insufficiency fractures occurred due to anatomical inferiority of bone tissue with unaccustomed or excessive loading of the residual limb in the prosthesis.

## Ethics Approval

The study was approved by the Bioethical Committee of Scientific and Research Institute of Rehabilitation of National Pirogov Memorial Medical University (Approval number: 05/2024, 07.02.2024), Vinnytsia, Ukraine.

## Consent for Publication

Written informed consent was provided by the patients to have the case details and any accompanying images published.

## Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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## Disclosure

The authors report no conflicts of interest in this work.

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