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Effect of implant surface nanotopography on neutrophil extracellular trap formation: An *in vitro* study

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Abstract:
Excessive neutrophil extracellular trap (NET) formation during the early healing phase around dental implants may impair peri-implant tissue integration, yet the influence of implant surface nanotopography on NET formation remains unclear. Therefore, it is of interest to compare the isolated human neutrophil responses to five titanium implant surface modifications (machined smooth, micro-rough, nanoporous, nanotubular and nano-hydrophilic) using extracellular DNA release, myeloperoxidase-DNA complexes, citrullinated histone H3 immunofluorescence, neutrophil elastase activity, reactive oxygen species production and cell viability. Extracellular DNA release was highest on machined smooth titanium (154.6 ± 21.8 ng/mL) and lowest on nano-hydrophilic titanium (86.9 ± 15.7 ng/mL), with significant differences in NET markers, elastase activity and ROS levels among groups ($p<0.001$). Nano-hydrophilic and nanotubular titanium surfaces significantly reduced NET formation without compromising short-term neutrophil viability. Thus, data shows that implant surface nanotopography can beneficially modulate the early inflammatory response and may enhance peri-implant healing by limiting excessive NET formation.

Keywords: Dental implant, titanium, nanotopography, neutrophil extracellular traps (NETs), NETosis, osseointegration, *in vitro* study

Background:
The ability to predictably achieve osseointegration and control early host response are critical to the success of dental implants. The initial biological response to implant placement is not bone formation, but rather the interaction between the titanium surface, blood proteins, platelets and innate immune cells. This initial interface will affect the composition of the provisional matrix and affect subsequent macrophage activity, angiogenesis, osteoblast recruitment and bone-to-implant contact. The long-term clinical success criteria focus on the lack of mobility, pain, progressive radiolucency and uncontrolled marginal bone loss, but some of these criteria are dependent on molecular and cellular events that occur within the first hours after placement **[1]**. Surface topography of titanium is therefore a key research area in implant biomaterials. Moderately rough surfaces tend to promote osteogenic cell attachment and early anchorage to bone better than smoother surfaces and nanoscale features provide an additional level of biological control through effects on protein adsorption, focal adhesion formation, cytoskeletal organization and immune-cell signaling **[2]**. The nanoscale range is especially important because many proteins in the extracellular matrix, integrins and clusters of membrane receptors function at dimensions less than 100 nm. Thus, the surface nanopores, nanotubes, nanonodules and hydrophilic nanostructures can influence host responses before cells start to spread on the implant surface **[3]**. The current surface development of implants has shifted from mechanical roughening to micro-nano hybrid surfaces designed to control osteogenesis, bacterial adhesion and immune resolution at the same time. It has been showed that well-defined nanotopography can influence cellular and molecular processes involved in osseointegration and nanostructured titanium surfaces can create a more favorable osteoimmune microenvironment **[4]**. Along with osteoblast and macrophage responses, there is a growing focus on neutrophils as one of the first inflammatory cells to reach a biomaterial surface following surgical implantation. The neutrophils are classically considered to be short-lived antimicrobial cells but they also control sterile inflammation, cytokine production, protease activity and recruitment of immune cells. One of the main neutrophil effector mechanisms is the generation of neutrophil extracellular traps (NETs) that are extracellular chromatin coated with granular proteins including myeloperoxidase and neutrophil elastase **[5]**. NETs can immobilize microorganisms and focus antimicrobial molecules at the site of infection, but excessive or persistent NET formation can exacerbate tissue injury, impair inflammatory resolution and increase proteolytic damage **[6]**. The biological significance of NET formation around implants is, therefore, complex: a controlled NET release can be beneficial for antimicrobial defense, while an uncontrolled NETosis can be a factor in the early development of peri-implant inflammation. The material microenvironment has been increasingly identified as a key regulator of neutrophil behavior. Neutrophil spreading, generation of reactive oxygen species, cytokine release and

NETosis are affected by surface chemistry, hydrophilicity, stiffness and roughness **[7]**. *In vitro*, hydrophilic titanium surfaces have been shown to have a decreased pro-inflammatory neutrophil response and decreased NET formation, indicating that modified implant surfaces may have a less destructive early innate immune response **[8]**. Likewise, mechanical signals like biomaterial stiffness can trigger NET formation via adhesion-dependent signaling, suggesting that neutrophils are not only responding to soluble inflammatory signals, but also to physical properties of biomaterials **[9]**. Therefore, it is of interest to compare the ability of human neutrophils to form NETs when stimulated by various nanotopographies of titanium implants.

Materials and Methods:

Study design and setting:

This *in vitro* experimental study was performed in the Department of Periodontology and Implant Biomaterials Research Laboratory for six months. The study assessed NET formation of human peripheral blood neutrophils cultured on titanium discs having five different surface configurations. The experimental unit was the surface of the titanium disc in contact with the standardized suspension of neutrophils. Each assay was repeated three times for each donor and surface group and the average of the three readings was used for statistical analysis.

Sample size estimation:

The difference in the primary outcome, extracellular DNA release, between the smooth and nano-hydrophilic titanium surfaces was expected to be 35 ng/mL and the pooled standard deviation was 18 ng/mL in a pilot experiment. A minimum of eight independent donor samples was needed with an alpha error of 0.05 and a power of 90%. Neutrophils from 10 healthy volunteers were added to compensate for the potential for variation in cell yield. For the primary extracellular DNA outcome, 50 donor-surface comparisons and 150 technical assay wells were tested for 5 surface groups.

Preparation and grouping of titanium discs:

Commercially pure grade IV titanium discs of 10 mm diameter and 2 mm thickness were used. The discs were divided into five groups: Group I, smooth titanium; Group II, micro-rough sand blasted and acid etched titanium; Group III, nanoporous surface of titanium oxide; Group IV, anodized surface of titania nanotubular; and Group V, nano-hydrophilic micro-nano surface of titanium. Before cell culture, all discs were ultrasonically cleaned in acetone, ethanol and sterile distilled water, then dried under filtered air, individually packed and gamma-irradiated.

Surface characterization:

Representative discs in each group were characterized prior to cell culture. Optical profilometry was used to evaluate the surface roughness parameters and the three-dimensional surface morphology was captured by scanning electron microscopy. Wettability was estimated by measuring the static water contact angle by the sessile drop method. Descriptive correlation with NET markers was made with mean arithmetic roughness, nanoscale feature diameter and contact angle.

Neutrophil isolation:

Peripheral venous blood was drawn from 10 healthy adult volunteers (22-35 years) who gave written informed consent. Smokers, individuals with systemic inflammatory disease, those requiring active periodontal treatment, diabetics, those who had had a fever, antibiotics or corticosteroid within the past 30 days, pregnant women and those who used anti-inflammatory medications regularly were excluded. Blood was collected in heparinized tubes and processed within one hour of collection.

Isolation and culture of neutrophils:

Human neutrophils were isolated by density gradient centrifugation and red blood cell lysis. The purity of the cells was confirmed by Wright-Giemsa staining and was accepted if the neutrophils were more than 95% pure. Viability was determined by trypan blue exclusion and only preparations with viability greater than 97% were used. Neutrophils were suspended in RPMI-1640 medium with 1% heat-inactivated fetal bovine serum and seeded on titanium discs in 24-well culture plates at a density of 5 × 10^5 cells per disc. The cells were cultured for 4 hours at 37°C in 5% CO_2. Phorbol 12-myristate 13-acetate treated wells were used as positive assay controls and neutrophils grown on tissue culture plastic without stimulant were used as baseline controls.

Assessment of NET formation:

Extracellular DNA in culture supernatants was measured by a fluorescent nucleic acid dye assay and reported as ng/mL. MPO-DNA complexes were quantified by enzyme-linked immunosorbent assay (ELISA) with anti-myeloperoxidase as capture antibody and anti-DNA as detection antibody and the results were expressed as optical density (OD) at 450 nm. Immunofluorescence staining was used to determine the expression of citrullinated histone H3. Fluorescence microscopy was used to photograph five random high power fields per disc and the percentage of citrullinated histone H3-positive extracellular web-like structures was determined by image-analysis software. The activity of neutrophils was measured in the supernatant using a colorimetric substrate assay and expressed as mU/mL.

Reactive oxygen species and viability assessment:

Generation of intracellular reactive oxygen species was detected by dichlorofluorescein diacetate (DCF-DA) fluorescence and expressed as relative fluorescence units. Neutrophut viability was determined after 4 hours of surface exposure by resazurin-based viability assay and expressed as percentage viability compared to unstimulated cells. These endpoints were added to see if the reduced NET formation was due to immunomodulation, rather than to generalized cytotoxicity.

Data analysis:

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Data was analyzed using statistical analysis which was implemented in SPSS version 26.0. The Shapiro-Wilk test was used to determine normality. Data for continuous variables are reported as mean ± SD. One-way analysis of variance (ANOVA) followed by Tukey post-hoc testing was used for intergroup comparisons for normally distributed variables. Kruskal-Wallis and Dunn post-hoc tests were used for non-parametric data. Pearson or Spearman correlation was used to determine the correlation between contact angle, roughness and NET markers as appropriate. The p value < 0.05 was regarded as statistically significant.

Results:

There was no visible damage to any of the titanium discs after sterilization and cell culture. Neutrophil purity was between 95.6% and 98.8% and baseline viability prior to seeding was 98.4 ± 1.1%. The roughness and wettability profile of the five surfaces were different. The surface roughness of the machined smooth group was the lowest and the contact angle was the highest, the nano-hydrophilic group had the lowest contact angle and the micro-nano surface morphology. The nanotubular group exhibited organized tubular nanofeatures with a mean diameter of 74.8 ± 8.6 nm. There were significant differences in the markers for NET between the surface groups. Extracellular DNA release was highest in the machined smooth group (154.6 ± 21.8 ng/mL), followed by the micro-rough group (136.2 ± 19.4 ng/mL), nanoporous group (112.5 ± 17.6 ng/mL), nanotubular group (98.3 ± 16.2 ng/mL) and nano-hydrophilic group (86.9 ± 15.7 ng/mL). The overall difference between the groups was statistically significant ($p < 0.001$). The results of post-hoc analysis revealed that the groups of nanotubular and nano-hydrophilic had significantly less extracellular DNA release than the machined smooth and micro-rough groups. MPO-DNA complexes and citrullinated histone H3-positive NET structures were similar. The lowest MPO-DNA optical density (0.41 ± 0.08) and citrullinated histone H3-positive area (12.6 ± 4.2%) were observed in the nano-hydrophilic group. The activity of neutrophils' elastase and generation of reactive oxygen species was also significantly decreased on nanotubular and nano-hydrophilic surfaces. In all titanium surface groups, the short-term cell viability was above 90% and there was no statistically significant difference in cell viability between the nanostructured groups and machined smooth titanium. A positive correlation was found between water contact angle and extracellular DNA release ($r = 0.71$, $p = 0.002$) by correlation analysis, suggesting that more NETs are formed on less hydrophilic surfaces. There was a moderate positive correlation between reactive oxygen species and MPO-DNA complexes ($r = 0.68$, $p = 0.004$). The correlation between surface roughness and NET markers was less strong than that of wettability and nanoscale feature organization **(Table 1-3).**

Table 1: Surface characterization of titanium disc groups

Group	Surface type	Mean roughness Ra (μm)	Dominant nanoscale feature	Water contact angle (°)
I	Machined smooth	0.18 ± 0.04	Linear machining grooves	82.4 ± 6.3
II	Micro-rough SLA-like	1.72 ± 0.21	Irregular micro-pits	68.7 ± 5.8
III	Nanoporous TiO2	0.96 ± 0.13	Nanopores 38.5 ± 6.4 nm	54.2 ± 4.9
IV	Titania nanotubular	1.08 ± 0.16	Nanotubes 74.8 ± 8.6 nm	43.6 ± 5.1
V	Nano-hydrophilic micro-nano	1.21 ± 0.18	Hydrophilic nano-network	18.9 ± 3.7

Table 2: Comparison of NET formation markers across implant surface groups

Group	Extracellular DNA (ng/mL)	MPO-DNA OD450	CitH3-positive NET area (%)	Neutrophil elastase (mU/mL)	p-value
Machined smooth	154.6 ± 21.8	0.78 ± 0.11	31.4 ± 6.8	48.9 ± 7.6	<0.001
Micro-rough SLA-like	136.2 ± 19.4	0.69 ± 0.10	27.8 ± 5.9	43.1 ± 6.8	
Nanoporous TiO2	112.5 ± 17.6	0.55 ± 0.09	19.7 ± 5.1	35.7 ± 5.9	
Titania nanotubular	98.3 ± 16.2	0.48 ± 0.08	15.9 ± 4.7	30.4 ± 5.2	
Nano-hydrophilic micro-nano	86.9 ± 15.7	0.41 ± 0.08	12.6 ± 4.2	26.8 ± 4.9	

Table 3: Reactive oxygen species generation, cell viability and post-hoc interpretation

Group	ROS (relative fluorescence units)	Cell viability after 4 h (%)	Main post hoc finding
Machined smooth	842.5 ± 96.3	93.8 ± 3.1	Higher NET markers than all nanostructured groups
Micro-rough SLA-like	766.4 ± 88.7	94.5 ± 2.8	Higher DNA and MPO-DNA than nanotubular and nano-hydrophilic groups
Nanoporous TiO2	621.7 ± 74.5	95.1 ± 2.9	Intermediate NET response
Titania nanotubular	548.2 ± 66.9	95.8 ± 2.6	Lower NET response than machined and micro-rough surfaces
Nano-hydrophilic micro-nano	492.6 ± 61.4	96.2 ± 2.4	Lowest NET and ROS markers without cytotoxicity

Discussion:

In this *in vitro* study, it was shown that the surface nanotopography of implants had a significant effect on the formation of NETs by human neutrophils. The smooth titanium machined surface had the highest NET-related response, while the nano-hydrophilic titanium had the lowest response. Nanotubular and nano-hydrophilic surfaces decreased extracellular DNA release, MPO-DNA complexes, citrullinated histone H3-positive NET structures, neutrophil elastase activity and generation of reactive oxygen species without affecting short-term cell viability. The results indicate that surface nanoscale organization and wettability can modulate early activation of neutrophils at the implant interface, rather than just suppress them in a non-specific manner. The early implant environment is biologically dynamic. Blood proteins quickly bind to titanium, platelets become activated and neutrophils move towards provisional matrix. Recent studies have shown that the early inflammatory-cell response to titanium and

zirconia surfaces can be influenced by surface roughness, chemistry and wettability, yet they also underscore the need for standardized outcome measures in implant biomaterial studies that are specific to NETs **[10]**. The present results corroborate the hypothesis that nanoscale design is not only an osteogenic variable, but also an immunomodulatory variable. Surface nanofeatures can affect the adsorption pattern of fibrinogen, albumin, complement proteins and immunoglobulins, which in turn can affect the ligands presented to neutrophil receptors. *In vivo* biomaterial evidence has also showed that controlled NETosis at the implant interface can promote inflammatory resolution and enhance bone formation on titanium implants, which further highlights the biological relevance of controlled NETosis at the implant interface **[11]**. The lower NET response observed in nano-hydrophilic titanium is clinically relevant as excessive NETosis can contribute to tissue damage via extracellular histones, proteases, oxidants and sustained inflammatory signaling. Peri-implant health is characterized by a balanced host response, while peri-implant diseases are characterized by chronic inflammation around implant-supported tissues and the progressive destruction of the supporting tissues **[12]**. A surface that does not trigger unnecessary sterile inflammatory amplification in the early healing period may then be beneficial, especially in patients with greater inflammatory susceptibility. Foreign body responses to biomaterials are not passive events but instead are orchestrated cellular events that include protein adsorption, recruitment of neutrophils, activation of monocytes/macrophages, release of cytokines and tissue remodeling. Chronic inflammation and fibrous encapsulation can result if the biomaterial-host interface does not resolve **[13]**. The early peri-implant wound is similar to the dental implant in that it is not encapsulated, but the same principles of controlled inflammation are at play. The decrease in MPO-DNA complexes and citrullinated histone H3-positive structures in the nano-hydrophilic group suggests that the extracellular DNA decrease was not just due to the decrease in cell numbers or assay artifact. This distinction is significant as necrosis, apoptosis and NETosis can all result in extracellular DNA. The current design features cell viability >90% and reduced MPO-DNA and citrullinated histone H3 staining, which indicate a genuine reduction in NET-associated activation. The interaction between neutrophils and the behavior of the subsequent macrophages is especially important. Macrophage activation profiles can be altered by the topography and wettability of titanium and polarization of macrophages is a key factor in determining the progression of early inflammation to repair or persistent tissue damage **[14]**. A surface that inhibits excessive NETosis will indirectly affect the macrophage phenotype by decreasing exposure to extracellular chromatin, histones, elastase and oxidative mediators. The NET profile was not completely accounted for by surface roughness. The micro-rough surface also showed higher roughness than the machined group, but it also showed significant NET formation. In contrast, moderate roughness resulted in lower NET markers on nanotubular and nano-hydrophilic surfaces. This means that the surface energy, nanoscale architecture and micro-scale roughness should be taken into account. In the context of reviews of surface modification of implants, it is also noted that the combination of topographical, chemical and wettability features influence osseointegration and the behavior of the biofilm, not just a single surface parameter **[15]**. This is also true for the reduction of ROS on nanotubular and nano-hydrophilic surfaces. ROS are part of multiple pathways of NETosis and can exacerbate local tissue damage if generated in excess. In the current study, generation of reactive oxygen species was associated with MPO-DNA complexes, which further suggests that oxidative activation played a role in NET formation. This is in line with the overall principle that integration of implants is dependent on controlled foreign-body reaction and not on the lack of inflammation **[16]**. The results also have implications for the prevention of peri-implant disease. Peri-implantitis is defined as the presence of inflammation around dental implants associated with progressive loss of supporting bone and is a result of both the presence of microorganisms and host inflammatory susceptibility **[17]**. A surface that helps to minimize excessive sterile NET release alone may not be sufficient to prevent biofilm-related disease, but it may help to create a more balanced early host response and may help to limit the risk of unchecked inflammation after placement. The ideal surface should not be immunologically inert, but rather should direct inflammation to a timely resolution. There are a number of limitations in the present study. First, it employed isolated neutrophils in a controlled *in vitro* environment, which could not simulate the entire peri-implant environment, in which platelets, complement, monocytes, endothelial cells, osteogenic cells, saliva and oral microorganisms interact simultaneously. Second, the exposure time was short (4 hours) and not chronic inflammation. Third, donor variability was limited by repeated testing of all surfaces, but only healthy young adults were tested. Fourth, standardized titanium discs allow for controlled assays, but do not simulate threaded implant macrogeometry. Whole-blood stimulation, bacterial biofilm challenge, macrophage-neutrophil co-culture and implant-shape replication should be added to future models. The present findings suggest a mechanism for NET formation, namely through cell adhesion. The spreading of the cells or engagement of the receptors can regulate NETosis and the extent of cell spreading or receptor engagement can affect the probability of chromatin externalization **[18]**. Recent evidence indicates that implant surface topography is not merely a structural feature but an active biological regulator of osteoblast, osteoclast, macrophage and neutrophil behavior, thereby influencing osseointegration and bone regeneration **[19]**. Biomimetic micro-/nanostructured and hierarchical surface topographies may improve cellular attachment, osteogenic differentiation, immune modulation and bone–implant integration, although these responses depend on surface roughness, feature scale, hydrophilicity and the surrounding biological microenvironment **[20]**. Neutrophil extracellular traps add an important early immunological dimension to implant healing, as balanced NET formation may support antimicrobial defence, whereas excessive NETosis may prolong inflammation and compromise regenerative healing,

highlighting NET regulation as a potential therapeutic strategy **[21]**. The conformational changes induced by the nano-hydrophilic and nanotubular architectures can be enough to decrease the release of NETs without compromising the viability of neutrophils. In general, these findings indicate that surface nanotopography is a relevant factor that can modulate neutrophil function and could be used to direct the development of future dental implant surfaces that promote both osseointegration and inflammatory resolution.

Conclusion:

Nano-hydrophilic and nanotubular implant surfaces significantly reduced NET formation compared with smooth and micro-rough titanium surfaces. Nano-hydrophilic titanium showed the most favorable immunomodulatory response, with lower NET activity and greater neutrophil viability. Thus, nanoscale surface design and wettability may help regulate early peri-implant inflammation, although further *in vivo* studies are required.

References:

[1] Albrektsson T *et al. Int J Oral Maxillofac Implants.* 1986 **1:**11 [PMID: 3527955]
[2] Wennerberg A & Albrektsson T. *Clin Oral Implants Res.* 2009 **20:**172 [PMID: 19663964]
[3] Variola F *et al. Nanoscale.* 2011 **3:**335 [DOI: 10.1039/c0nr00485e]
[4] Karazisis D *et al Int J Nanomedicine.* 2016 **11:**1367 [DOI: 10.2147/IJN.S101294]
[5] Ma QL *et al. Biomaterials.* 2014 **35:**9853 [PMID: 25201737]
[6] Brinkmann V *et al. Science.* 2004 **303:**1532 [PMID: 15001782]
[7] Papayannopoulos V. *Nat Rev Immunol.* 2018 **18:**134 [PMID: 28990587]
[8] Abaricia JO *et al. Biomater Sci.* 2020 **8:**2289 [PMID: 32163073]
[9] Abaricia JO *et al. Biomaterials.* 2021 **271:**120715 [PMID: 33677375]
[10] Elangovan G *et al. Clin Exp Dent Res.* 2022 **8:**818 [PMID: 35535662]
[11] Morandini L *et al. Acta Biomater.* 2023 **166:**670 [PMID: 37187302]
[12] Berglundh T *et al. J Clin Periodontol*. 2018 45:S291 [PMID: 29926491]
[13] Anderson JM *et al. Semin Immunol*. 2008 **20:**86 [PMID: 18162407]
[14] Hotchkiss KM *et al. Acta Biomater*. 2016 **31:**425 [PMID: 26675126]
[15] Kligman S *et al. J Clin Med.* 2021 **10:**1641 [PMID: 33921531]
[16] Trindade R *et al. Clin Implant Dent Relat Res*. 2016 **18:**192 [PMID: 25257971]
[17] Schwarz F *et al. J Clin Periodontol.* 2018 **45:**S266 [PMID: 29926484]
[18] Lelliott PM *et al. Immunohorizons.* 2022 **6:**170 [PMID: 35193943]
[19] Guo W *et al. Mater Today Bio.* 2025 **34**:102066. [PMID: 40735704]
[20] Berger MB *et al. Biomimetics (Basel).* 2022 **7**:46. [PMID: 35466263]
[21] Al-Ghurabi BH *et al. Middle East Res J Dent.* 2024 **4**:26. [DOI: 10.36348/merjd.2024.v04i04.001]