京都大学	博士（　医　学　）	氏　名	LI　YUANJIAOZI (李 媛姣子)
論文題目	Modified gelatin hydrogel nonwoven fabrics (Genocel) as a skin substitute in murine skin defects （ マウス皮膚欠損創における改良型 Genocel の新規人工真皮としての有用性 ）		

（論文内容の要旨）

[Introduction] The skin functions as a vital protective barrier against injuries and harmful agents, and defects resulting from trauma or tumor resection can lead to bacterial invasion and body fluid loss. Skin substitutes offer a solution by creating a temporary barrier between the external and internal body environments. Previously, the potential of Genocel (NIKKE MEDICAL Co., Ltd., Osaka, Japan), a scaffold for three-dimensional cell culture, as a new skin substitute was explored. Genocel, gelatin hydrogel nonwoven fabrics, is manufactured using the solution-blow method, demonstrating high mechanical strength to maintain a porous structure that enables cells to infiltrate and distribute homogeneously. However, when applied to skin defects in mice, Genocel showed advantages in capillary formation but failed in granulation tissue formation and macrophage infiltration compared to the conventional skin substitute Pelnac. This inferiority could be attributed to the rapid degradation of Genocel, as it was not originally developed for use as a skin substitute.

[Materials and Methods] To optimize Genocel, the manufacturing process was modified to decrease its water content, thereby prolonging degradation time, and to assess its efficacy as a skin substitute in the healing process. Genocel with a low water content (Genocel-L) was prepared and the difference in water content compared to that of the conventional Genocel was confirmed. Degradation tests were performed using collagenase and compared among Genocel-L, Genocel, and Pelnac sheets. In the *in vivo* study, sheets of Genocel-L or Pelnac were applied to skin defects created on the backs of C57BL/6JJcl mice. On days 7, 14, and 21, the remaining wound area was evaluated and specimens were harvested for Hematoxylin and Eosin, Azan, anti-CD31, CD68, and CD163 staining to assess neoepithelialization, granulation tissue formation, angiogenesis, and macrophage infiltration.

[Results] Genocel-L has a lower water content compared to the conventional Genocel and degrades more slowly than both Genocel and Pelnac. In the *in vivo* experiment, no significant differences were observed between Genocel-L and Pelnac in relation to the wound area, neoepithelial length, granulation formation, and the number of newly formed capillaries. The area of newly formed capillaries in the Pelnac group was significantly larger than that in the Genocel-L group on day 21 ($p<0.05$). Regarding macrophage infiltration, significantly more M2 macrophages were induced in the Pelnac group on days 14 and 21, and the M2 ratio was larger in the Pelnac group ($p<0.05$) during the entire process.

[Conclusions] Genocel-L has a lower water content and longer degradation time compared to the conventional Genocel. Genocel-L had equivalent efficacy as a skin substitute to Pelnac, and can therefore be considered feasible for use as a skin substitute. However, a manufacturing method that can further modify Genocel-L is required to recover its early angiogenic potential.

（論文審査の結果の要旨）

Genocel はゼラチンハイドロゲルから成る不織布であり、湿潤した状態でも三次元構造を維持することができ、栄養物質や酸素の取り込み、代謝産物の排出を含む物質交換に有利であるため三次元細胞培養に使用されている。このことから、全層皮膚欠損創に貼付し真皮再生に使用される人工真皮として応用可能と考えている。過去の研究では、Genocel は早期の血管新生を誘導するが、肉芽組織の形成は不十分であった。

本研究では、Genocel の肉芽組織形成を改善することを目的とし、改良型 Genocel を作成し、肉芽形成を含む創傷治癒効果を検討した。改良型 Genocel では、従来型よりも熱架橋を強化した。マウス皮膚欠損創モデルを用いて改良型 Genocel と既存の人工真皮製品（ペルナック®）とを比較検討した。改良型 Genocel は生体分解性の延長、含水率の低下を示し、ペルナックと同程度の肉芽組織が可能であった。また、改良型 Genocel は創閉鎖、新生上皮形成、血管形成、マクロファージ誘導などにおいても、ペルナックと同等の結果を示した。一方で、改良型 Genocel は早期の血管新生を誘導する優位性を失ったため、今後、さらなる改良を加える必要があると考えられた。

以上の研究は改良型 Genocel の人工真皮としての臨床応用の可能性を示唆し、今後の製品化を含めた医療機器の開発に寄与するところが多い。

したがって、本論文は博士（　医学　）の学位論文として価値あるものと認める。

なお、本学位授与申請者は、令和 6 年 2 月 15 日実施の論文内容とそれに関連した試問を受け、合格と認められたものである。

要旨公開可能日：　　年　　月　　日　以降