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Advances in Machine Learning and Deep Learning Applications for Intracranial/Extracranial Atherosclerotic Plaques and White Matter Hyperintensities

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Abstract

Machine learning (ML) and deep learning (DL), as pivotal components of artificial intelligence (AI), are revolutionizing precision medicine through their robust learning capabilities and image recognition functions. These technologies have significantly impacted disease diagnosis, therapeutic evaluation, prognosis prediction, and survival analysis. This review synthesizes recent advances in ML and DL applications for intracranial/extracranial atherosclerotic plaques and white matter hyperintensities (WMH), while critically analyzing current challenges and future directions.

Keywords: machine learning, deep learning, atherosclerosis, white matter hyperintensities, HR-MR VWI

1. Introduction

Intracranial/extracranial atherosclerotic plaques and white matter hyperintensities (WMH) are critical imaging biomarkers of cerebrovascular diseases. Accurate assessment of these markers is essential for diagnosis, treatment planning, and prognosis prediction. While high-resolution magnetic resonance vessel wall imaging (HR-MR VWI) enables detailed plaque characterization and lumen-wall visualization, traditional manual interpretation suffers from subjectivity and inefficiency. Recent advancements in AI, particularly ML and DL, offer automated solutions for plaque and WMH analysis through superior image recognition and high-dimensional feature extraction. However, existing studies often focus on isolated tasks, lacking systematic integration. This review comprehensively evaluates ML and DL applications in atherosclerotic plaques and WMH research, identifies current limitations, and outlines future directions.

2. Overview of Machine Learning and Deep Learning

The medical field is undergoing a transformative shift in diagnostic and therapeutic paradigms driven by artificial intelligence (AI), with particularly remarkable advancements in automated image analysis and pathological feature identification. Based on differences in modeling principles, machine learning (ML) can be categorized into three paradigms: supervised learning, unsupervised learning, and reinforcement learning (Srinivas & Young, 2023). The foundational theory of ML involves data-driven optimization of model parameters to minimize errors between predicted outputs and ground-truth results (Chen et al., 2017; Yan & Wang, 2022). Supervised learning, the most mature paradigm in clinical diagnostics, has significantly improved the precision of tumor staging assessments and the efficacy of non-invasive detection. Unsupervised learning (Matteucci et al., 2024), which utilizes unlabeled data to construct analytical models, exhibits unique advantages in disease subtype clustering and association rule mining, though it faces limitations in predictive stability. Reinforcement learning (Xuan et al., 2022) employs dynamic decision-making mechanisms, using delayed feedback to optimize agent strategies, with its core principle lying in balancing the exploration of new pathways and the exploitation of existing knowledge.

As a pivotal advancement in ML, deep learning (DL) simulates human cognitive mechanisms through biomimetic neural network architectures. By leveraging backpropagation algorithms, DL achieves hierarchical feature abstraction and demonstrates exceptional performance in medical pattern recognition tasks (Lin, 2023; Thompson et al., 2020; Wagner et al., 2021). Among DL architectures, convolutional neural networks (CNNs) have achieved breakthroughs in medical image classification and lesion segmentation (Li et al., 2022; McBee et al., 2018; Wang et al., 2023). Radiomics technology further enhances diagnostic objectivity by integrating quantitative feature analysis with ML algorithms. This approach automates the extraction of multidimensional parameters from regions of interest and constructs auxiliary diagnostic systems through statistical modeling, thereby significantly improving disease classification and severity assessment (Hatt et al., 2019; Lambin et al., 2012; Mayerhoefer et al., 2020). Recent advances (Y.-F. Chen et al., 2023) highlight that hybrid models combining DL and radiomics enable precise automated identification and quantitative analysis of carotid plaques, offering innovative solutions for vascular pathology evaluation.

Notably, DL is evolving toward large-scale models supported by massive datasets and computational power. The Transformer architecture overcomes limitations in sequence modeling through attention mechanisms, diffusion generative models are emerging in medical image synthesis, and pretrained models such as GPT and BERT have pioneered new pathways for cross-modal medical data analysis. These technological breakthroughs continue to drive the profound integration of AI into healthcare applications.

3. Applications of Machine Learning and Deep Learning in Intracranial/Extracranial Atherosclerosis

3.1 Plaque Segmentation and Feature Assessment

Current imaging modalities for detecting intracranial and extracranial atherosclerotic plaques include ultrasonography, computed tomography angiography (CTA), and high-resolution magnetic resonance vessel wall imaging (HR-MR VWI). Although ultrasonography demonstrates high sensitivity in identifying carotid plaques and detecting hemodynamic alterations, its utility in intracranial artery evaluation remains limited due to acoustic shadowing from the skull, insufficient penetration depth for assessing deep vessels, and challenges in collateral circulation evaluation. CTA effectively evaluates lumen stenosis and calcified components but lacks precision in characterizing small calcified or non-calcified plaques, with further limitations in assessing plaque composition and vulnerability. In contrast, HR-MR VWI, with its superior spatial resolution, provides unique advantages for morphological measurements of intracranial/extracranial arterial walls and quantitative analysis of plaque composition (Saba et al., 2018). However, traditional manual interpretation exhibits significant limitations: Operators require specialized expertise in atherosclerotic pathology and extensive clinical experience, results are susceptible to inter-operator variability in expertise and subjective interpretation, manual delineation suffers from high inter-observer variability and time inefficiency. Recent advancements in artificial intelligence (AI), particularly machine learning (ML) and deep learning (DL), offer novel solutions for plaque analysis. These technologies leverage existing data to extract high-dimensional features, construct diagnostic and predictive models, and significantly enhance the accuracy and efficiency of plaque evaluation and cerebrovascular event prediction.

DL-based automated analysis systems standardize image segmentation workflows through feature learning from large-scale annotated datasets, markedly improving processing efficiency and result consistency. Current research on HR-MR VWI image analysis focuses on algorithmic architecture innovation and multi-model synergy. For instance: Wan et al. (Wan et al., 2022) developed a 3D convolutional network system that achieves automatic vascular centerline tracking, geometric correction, and wall morphology parameter measurement, with segmentation accuracy exceeding 0.9 for intracranial/extracranial arteries. Shi et al. (F. Shi et al., 2019) pioneered the application of U-Net architecture for whole-brain HR-MR VWI segmentation, achieving Dice similarity coefficients of 0.89 (wall) and 0.77 (lumen). Their model successfully identified statistically significant differences in normalized plaque indices between symptomatic and asymptomatic groups across 24 severe stenosis cases. Wu et al. (Wu et al., 2019, 2024) proposed the DeepMAD multi-task framework, which excels in carotid plaque segmentation and pathological assessment, with performance further enhanced through joint optimization strategies.

3.2 Radiomics-Driven Quantitative Plaque Assessment

Radiomics-based quantitative plaque evaluation introduces a new dimension for risk stratification in ischemic cerebrovascular diseases. Pathological studies of acute ischemic stroke (AIS) reveal that atherosclerotic plaque morphology and composition are more effective predictors of cerebrovascular events than lumen stenosis severity (Prabhakaran et al., 2021; Tian et al., 2023). Current research paradigms employ ML algorithms to construct high-dimensional feature analysis models, enabling intelligent assessment of plaque heterogeneity. Quantitative analysis of anterior circulation plaques demonstrates that symptomatic cases exhibit significantly higher signal intensity distributions in histogram parameters compared to asymptomatic controls (Yu et al., 2019), a pattern also validated in posterior circulation plaque evaluation (Z. Shi et al., 2018). Shi et al. (Z. Shi et al., 2020) further demonstrated that histogram feature dispersion serves as a robust biomarker for discriminating culprit plaques, underscoring the superiority of quantitative imaging features in characterizing plaque heterogeneity. Notably, integrated models combining HR-MR VWI multimodal data with random forest algorithms have demonstrated diagnostic efficacy surpassing traditional methods in vulnerable plaque identification (Z. Shi et al., 2018). For culprit plaque discrimination, Zhang et al. (Zhang Guiling et al., 2023) optimized classification performance using extreme gradient boosting in a multi-sequence fusion model, providing technical support for precision diagnostics.

3.3 Cerebrovascular Events

Current research on the association between atherosclerosis and acute ischemic stroke (AIS) focuses on three dimensions: elucidation of pathogenic mechanisms, event risk prediction, and recurrence warning. The prevailing research paradigm involves integrating quantitative features from high-resolution vessel wall imaging (HR-VWI) with conventional imaging parameters to construct machine learning (ML)-based predictive frameworks. For instance: Li et al. (Li et al., 2023) developed an ensemble learning model that demonstrated superior performance in discriminating stroke mechanisms, achieving a 32% improvement in predictive accuracy compared to traditional models. Wang et al. (Wang et al., 2023) employed survival analysis to build a recurrence risk assessment system, identifying high-order texture features (e.g., gray-level co-occurrence matrix parameters) as optimal predictors of prognosis. Recent evidence (Tang et al., 2022) indicates that combining radiomic features with nomogram tools enhances stroke recurrence warning sensitivity to 92%, providing quantitative guidance for personalized treatment. The integration of deep learning (DL) and radiomics has established novel pathways for cerebrovascular event risk assessment. For example: Chen et al. (Y.-F. Chen et al., 2023) developed an intelligent diagnostic system using an object detection network to automate plaque component analysis, achieving 94.81% accuracy in AIS risk stratification. In image preprocessing, a transfer learning-based DL model achieved precise vascular wall segmentation (Song et al., 2023). This method, when combined with a support vector machine (SVM)-based multimodal diagnostic system, demonstrated exceptional performance in tumor grading, suggesting its potential applicability in cerebrovascular diseases.

Notably, AIS pathogenesis involves multidimensional factors, including plaque stability, hemodynamic alterations, and molecular biological regulation (Ajoolabady et al., 2021; AlRuwaili et al., 2024; Arul et al., 2023; Biose et al., 2023). Models relying solely on imaging features risk systemic bias due to incomplete representation of these complex interactions. Emerging proteomics studies (Theofilatos et al., 2023) have confirmed that multi-omics data integration models improve predictive performance by 19.7% compared to single-modality approaches.

4. Applications of Machine Learning and Deep Learning in WMH Research

4.1 WMH Segmentation

Accurate segmentation of white matter hyperintensities (WMH) is fundamental for investigating their pathological mechanisms and clinical correlations. Traditional manual segmentation, reliant on radiologists' expertise, is time-consuming, subjective, and suffers from poor reproducibility. Deep learning (DL) techniques have significantly enhanced segmentation efficiency and precision through automation. For instance: Dadar et al. (Dadar et al., 2017) compared 10 classification techniques for WMH segmentation and demonstrated that the random forest classifier achieved optimal performance on a dataset comprising T1-weighted imaging (T1WI), T2-weighted imaging (T2WI), proton density (PD), and fluid-attenuated inversion recovery (FLAIR) scans, with a Dice Kappa coefficient of 0.66 ± 0.17. One study (L et al., 2019) have proposed a context restoration-based self-supervised learning strategy for medical image analysis, which exhibited superior performance in classification, localization, and segmentation tasks across fetal ultrasound, abdominal CT, and brain MR images. Park et al. (Park et al., 2021) developed a multi-scale highlighted foreground U-Net for WMH segmentation, achieving the highest overall evaluation score, Dice similarity index, and F1-score in the MICCAI 2017 WMH Segmentation Challenge. Shan et al. (Shan et al., 2021) clinically validated a DL-based automated system for segmenting cerebral small vessel disease-related WMH (CSVD-WMH), which outperformed existing methods on both internal and external test sets. A study (S et al., 2024) combined deep neural networks with Transformer architectures for automated cervical cancer segmentation and survival prediction, demonstrating superior segmentation performance and significantly outperforming traditional methods in survival analysis.

4.2 WMH Quantification and Clinical Decision-Making

Cerebral small vessel disease (CSVD) is a common neurological disorder predominantly affecting elderly populations. Its pathophysiological mechanism primarily involves microvascular structures, including small arteries, arterioles, capillaries, venules, and small veins, leading to clinical manifestations such as cognitive impairment and vascular dementia. Due to the nonspecific clinical presentation, diagnosis currently relies heavily on neuroimaging. White matter hyperintensities (WMH) represent one of the hallmark imaging features of CSVD, alongside other radiological markers such as lacunar infarcts, cerebral microbleeds (CMBs), enlarged perivascular spaces (EPVS), recent small subcortical infarcts, brain atrophy, and cortical superficial siderosis (cSS) (Hu Wenli et al., 2021). Previous studies on WMH (Erten-Lyons et al., 2013; Pan et al., 2024; Williams et al., 2017; Zhai et al., 2020) have focused on associations between imaging features and clinical phenotypes, exploration of pathophysiological mechanisms, and prediction of cognitive outcomes and prognosis. However, these investigations often relied on cross-sectional designs, manual or semi-automated segmentation methods, and single-modality imaging analyses, resulting in limitations such as challenges in causal inference, high sample heterogeneity, inconsistent quantification standards, and insufficient biological mechanistic explanations.

The integration of radiomics and deep learning (DL) has provided robust tools for WMH quantification. For example: Shi et al. (Y. Shi et al., 2022) conducted a bibliometric analysis to elucidate the intellectual structure and emerging trends in WMH research, emphasizing its associations with cognitive impairment, stroke, and neuroimaging characteristics in CSVD. Rudie et al. (Rudie et al., 2019) employed a 3D

U-Net model to differentiate glioma-related abnormalities from WMH in brain MRI data, achieving a Dice coefficient of 0.42, thereby demonstrating DL's efficacy in processing multi-disease neuroimaging datasets. Rachmadi et al. (Rachmadi et al., 2018) proposed a convolutional neural network (CNN) framework incorporating global spatial information, which significantly improved segmentation accuracy by integrating spatial coordinates, yielding a mean Dice similarity coefficient of 0.5389. Huff et al. (Huff et al., 2021) focused on enhancing the interpretability of DL models, exploring visualization techniques such as Gradient-weighted Class Activation Mapping (Grad-CAM) and attention maps to decipher model decision-making processes. This advancement is critical for improving the transparency and clinical acceptability of WMH analysis. Collectively, these studies demonstrate that DL not only enhances WMH segmentation accuracy but also supports clinical decision-making through interpretability techniques, laying a solid foundation for future research and clinical applications of WMH.

4.3 Cognitive Impairment Disorders and Mechanistic Insights into WMH Pathology

Artificial intelligence (AI) has demonstrated significant value in predicting the association between white matter hyperintensities (WMH) and cognitive impairment. Feng et al. (Feng et al., 2025) employed VB-Nets, a deep learning convolutional neural network, to automatically identify and segment whole-brain subregions and WMH. By extracting radiomic features from WMH and bilateral hippocampal regions, they constructed a combined feature model to detect cognitive impairment in WMH patients. The 3D VB-Net algorithm exhibited strong performance in WMH segmentation (Dice = 0.789, lesion F1-score = 0.764). Additionally, studies indicate that combined analysis of WMH and Aβ-amyloid significantly enhances the predictive capability for cognitive impairment, particularly in early stages. For example, Lorenzini et al. (Lorenzini et al., 2022) evaluated regional associations between WMH and Aβ-amyloid across brain regions using PET imaging, revealing two distinct pathological patterns that significantly predict cognitive decline.

Another study (da Silva et al., 2023) elucidated the mechanistic link between WMH and information processing speed (IPS) deficits in cerebral small vessel disease (CSVD) by investigating cortical thinning and network disruption. By assessing the mediating roles of cortical thickness and structural/functional brain connectivity in the relationship between WMH and IPS, the study identified significant associations ($p < 0.05$) among WMH volume/location, cortical thickness, brain connectivity, and IPS performance in CSVD patients. Specifically, frontal cortical thickness, functional sensorimotor networks, and the posterior thalamic radiation were identified as critical mediators of the WMH-IPS relationship. These findings underscore the importance of multimodal imaging data in early cognitive impairment diagnosis. Future research should prioritize the development of multimodal causal inference models to clarify the temporal relationships and interactions between WMH, neurodegeneration, and vascular pathologies.

5. Conclusions and Future Perspectives

The integration of deep learning (DL), machine learning (ML), and radiomics has introduced transformative methodologies for rapid plaque characterization and cerebrovascular event risk stratification. Despite these advancements, significant challenges persist, such as limited model interpretability, ambiguous correlations between high-dimensional features and clinical characteristics, insufficient reproducibility, and constraints in data quality and algorithmic robustness. Furthermore, most studies rely on single algorithms trained on small, single-center datasets lacking external validation, which compromises model generalizability. To address these limitations, future efforts should prioritize the following strategies: Clinical translation, enhancing AI transparency and clinical adoption through visualization techniques (e.g., gradient heatmaps) and medical knowledge graphs. Standardization, establishing unified imaging protocols and data acquisition workflows via multi-center collaborations. Data infrastructure, developing large-scale databases for head and neck atherosclerosis to improve model generalizability. Multi-Omics integration, combining radiomics with genomics, pathomics, and clinical biomarkers to elucidate molecular mechanisms and advance personalized therapeutics.

Critical research directions include constructing multidimensional data fusion frameworks, leveraging HR-MR VWI radiomics to characterize plaque morphology, integrating proteogenomic data to decode molecular pathways, and establishing precision prediction models that bridge imaging, molecular, and clinical domains. In parallel, ML/DL applications in white matter hyperintensity (WMH) research have expanded from basic segmentation tasks to mechanistic exploration and clinical prediction. Nevertheless, challenges remain: Data heterogeneity-inconsistent standardization across multi-center WMH datasets necessitates international shared databases (e.g., ADNI, UK Biobank) to enhance algorithmic robustness. Interpretability, the "black-box" nature of DL models hinders clinical trust, this limitation can be mitigated by integrating attention mechanisms (e.g., Grad-CAM) to visualize critical lesion regions. Multimodal limitations, overreliance on MRI underscores the urgency to incorporate PET (e.g., amyloid imaging) and liquid biopsy data, enabling holistic pathological mapping of WMH.

By fostering technological innovation and interdisciplinary collaboration, AI-driven frameworks hold immense potential to revolutionize the precision diagnosis and treatment of cerebrovascular diseases and WMH, ultimately improving patient outcomes.

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From Energy Dissipation to Information Density: Metabolic Rate in the Geometry of Aging

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Abstract

We study the relationship between basal metabolic rate and weight throughout life in humans. Our previous work has yielded tables and graphs with specific data. In this article, we discuss its relationship to the geometry of biological systems. We also address its relationship to the holographic description of biological systems in general. This allows us to approach the fundamentals of the aging process through its determinants: the size, shape, and dimension of living beings.

Keywords: basal metabolic rate, relative surface, Bekenstein border, hologram, geometric phase, Margalef principle

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1. Introduction

The concept of *velocity* originates from physics and quantifies the distance traveled by an object over a given time interval, formalized as $v = d/t$. By analogy, if the distance traveled is replaced by the energy expended by an organism over a specific period, we obtain its *metabolic rate*.

However, when considering the energy expenditure of an organism (rather than the displacement of an object) over time, the scenario becomes more complex. This complexity arises due to the involvement of numerous additional variables beyond those in mechanical velocity. Below, we analyze the most significant factors.

Living organisms increase their mass and size—they grow—until a certain point in their lifespan (shortly after puberty in humans and other species). Consequently, the energy expended by the organism rises during a significant portion of its lifetime.

The observation that a physical system can dissipate increasing amounts of energy might mistakenly suggest a violation of the **second law of thermodynamics**. Yet, it is unsurprising that an adult human dissipates more total energy than an infant over the same period. Similarly, an elephant weighing 5 tons will inevitably dissipate more energy than a mouse weighing a few dozen grams in the same timeframe.

In both cases, the greater total energy dissipation is due to the system's larger mass. This does not imply a higher *velocity* of metabolic reactions but rather a higher *frequency* of those reactions (due to increased mass) occurring at the same intrinsic rate. Frequency and velocity are distinct concepts.

All physical systems must comply with the **second law of thermodynamics**, and the evolution of energy dissipation in such systems must be expressed consistently with this law.

One way to demonstrate the decline in energy transformation capacity is to express energy dissipation **per unit mass** (preferably dry or water-free mass, if possible) rather than per total system mass. This approach uses metabolically active mass as the reference unit.

Consequently, it becomes apparent that a mouse dissipates more energy per unit mass than an elephant, just as a human infant dissipates more energy per unit mass than an adult of the same species. This framework restores conceptual order.

Only then can a precise definition of **metabolic velocity** be established, as metabolic reactions occur within a given mass—just as distances are traversed over a given time interval.

The analysis of results from prior research by these investigators (Barragán, J. & Sánchez, S., 2022) yields particularly insightful conclusions concerning this issue

When comparing the total BMR/day with the dry BMR/Kg, R^2 has a value of 0.96 ($p < 0.02$), which is statistically significant as can be seen in Figure 1.

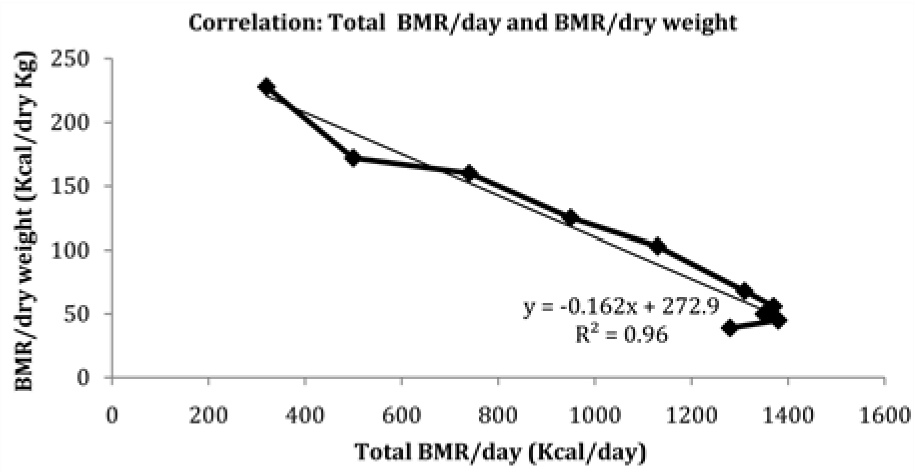

Figure 1. Relationship between total energy dissipation and energy dissipation per body mass unit

But when comparing the total BMR/day with the total body mass, R^2 has a value of 0.84 (NS), showing that there is no statistically significant association, as can be seen in Figure 2.

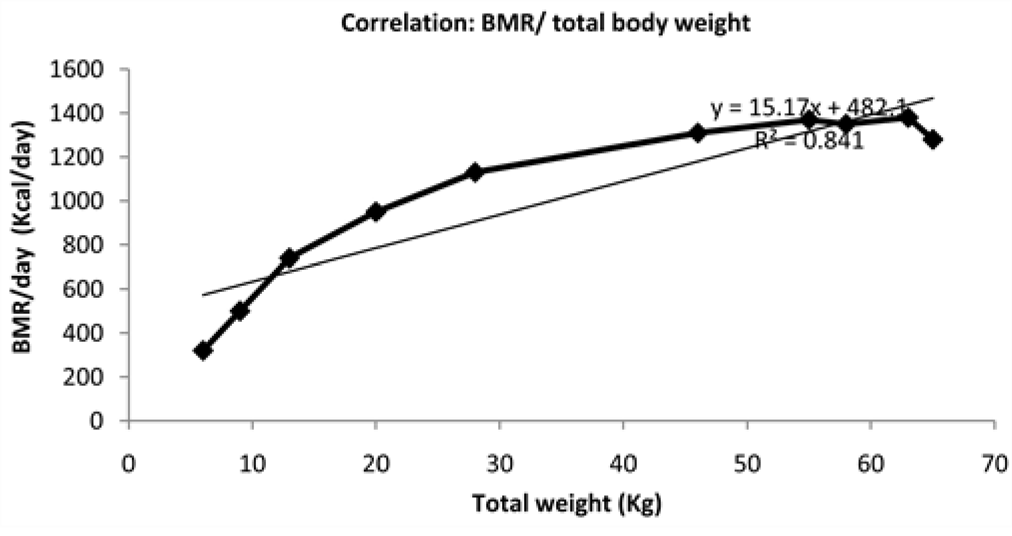

Figure 2. Relationship between total energy dissipation and the total body mass of the organism

These results are nothing more than the formalization of simple reasoning that arises from analyzing Table 1: If 6 kg of total body mass dissipates 320 Kcal/day, it would be expected that 65 kg of total body mass dissipates 3466 Kcal/day. However, that is not what happens. An older adult, weighing 65 kg, dissipates 1280 Kcal/day.

Table 1.

Age (years)	Total Weight (kg)	Total BMRl/day (Kcal/day)	BMR/kg (Kcal/dry weight)	Dry weight (kg)
0 - 0.5	6	320	228	1.4
0.5 - 1	9	500	172	2.9
1 - 3	13	740	160	4.6
4 - 6	20	950	125	7.6
7 - 10	28	1130	103	10.9
11 - 14	46	1310	68	19.3
15 - 18	55	1370	56	24.2
19 - 24	58	1350	50	26.6
25 - 50	63	1380	45	30.2
51 or more	65	1280	39	32.5

Table 1 shows total weight values, total kcal dissipated per day, and BMR/dry kg for different ages. Sample demographic characteristics: Argentine population white (Hispanic) race. Sample size: n = 10,960.

If instead, we observe the BMR/dry kg (fourth column) and the dry weight (fifth column) for each age, the following can be seen: A newborn whose dry BMR/kg is 228 Kcal and whose dry weight is 1.4 kg, dissipates 319.2 Kcal. An older adult, weighing 65 kg, whose BMR/dry kg is 39 Kcal and whose dry weight is 32.5 kg, dissipates 1267 Kcal/day. That is exactly what happens: a newborn dissipates 320 Kcal/day, and an older adult 1280 Kcal/day (third column of Table 1).

2. A Logical Equivalence

Continuing with the reasoning, if a tiny baby dissipates energy at a certain rate, and as it grows (increasing its mass) that rate decreases, then we have a metabolic acceleration. A negative acceleration.

In previous studies, the authors worked on this logical equivalence between mechanical velocity and metabolic velocity, as well as between mechanical acceleration and metabolic acceleration.

Of course, we are not proposing a physical equivalence between the two velocities, because we know they measure different variables and properties. But both concepts have the same truth value and the same rate of change. Their formalizations reveal the logical equivalence between the two.

It is important to know that we have metabolically accelerated systems. And to understand the origin of acceleration, we must analyze the calculation of energy dissipation in a biological physical system.

Another publication by the authors should be cited here to understand the problem. (Barragán, J. & Sánchez, S., 2023)

From the thermodynamic point of view, in a unicellular system we can calculate the total energy dissipated as the sum of the dissipations that occur in the system, in a similar way to the calculation of the linear momentum in an inertial system when we study mechanics (no fictitious forces are involved). (Ciufolini, I., 2007)

When analyzing the dissipation of energy, we will see that the system dissipates less and less energy (second law of thermodynamics), but it does so constantly generation after generation. Its increase in mass does not exceed the range of unity and the dissipation of energy per unit of mass is equal to the total dissipation of the system.

Yet when we study a complex system, the situation is different. The system dissipates more and more energy until it reaches its maximum complexity and also increases its mass above the value of unity. (Østbye, T., Malhotra, R. & Landerman, L.R., 2011; West, G.B. & Brown, J.H., 2004) What are the possible interpretations of this situation?

Serious thought should be given to the meaning of this question. In a complex biological system, self-organization operates as a fictitious force, which can be seen in the increase in mass of the system and in the apparent increase in its energy dissipation capacity. Self-organization operates as a particular force of aggregation of matter, which leads to the increasing ordering of it. (Isaeva, V.V., 2012; Wedlich-Söldner, R. & Betz, T. 2018; Ivanitskii, G.R., 2017)

Thus, the calculation of the total energy dissipated by the system does not depend only on the energy dissipated per unit of mass, but the intervention of the fictitious force of self-organization must be considered. It is a situation similar to that of non-inertial systems when we study mechanics (fictitious forces intervene when we calculate the linear momentum of the system). (Kamalov, T.F., 2010)

This implies that the decline in energy dissipation per unit mass in complex multicellular living things is not due to the second law of thermodynamics alone. The action of the self-organization force must also be considered.

The decline in energy dissipation per unit mass is not constant in the case of a complex multicellular organism. It behaves like a negative "metabolic acceleration" and because it is an accelerated system it turns out to be equivalent to what in mechanics is a non-inertial system. (Kamalov, T.F., 2010)

As demonstrated previously, the fundamental correspondence between mechanical and metabolic velocity metrics reduces to: (Barragán, J. & Sánchez, S., 2024; Barragán, J. & Sánchez, S., 2025)

We formalize the metabolic acceleration as Ma = BMR/m^2, where Ma is the metabolic acceleration; BMR is the energy dissipated, expressed in kcal per unit of mass $\frac{kcal}{m}$ and m is the mass expressed in kg of weight.

So the metabolic acceleration is $Ma = \frac{\frac{kcal}{m}}{m} = \frac{kcal}{m^2}$.

We formalize the mechanical acceleration as $A = \frac{d}{t^2}$ where A is the mechanical acceleration; d is the distance traveled per unit of time, expressed in meters per second; and t is the time expressed in seconds.

As the speed is $v = \frac{d}{t}$ where d is the distance expressed in meters and t is the time expressed in seconds.

Therefore, the mechanical acceleration is $A = \frac{\frac{m}{s}}{s} = \frac{m}{s^2}$.

We define this logical equivalence as

$$\frac{BMR}{m^2} :\Leftrightarrow \frac{d}{t^2}$$

3. When Do We Start to Age?

We define aging as the gradual loss of self-organization and homeostatic capacity. To understand how, when, and why aging occurs, we must first revisit the fundamentals of biological self-organization and homeostasis.

Living organisms are complex physical systems comprising: (Margalef, R., 2002; Margalef, R., 1995)

1) An energy-dissipating system, coupled with
2) A complementary energy-to-information recovery system

This dual architecture enables organisms to:

- Self-organize by generating structure from dissipated energy
- Maintain structural identity (homeostasis) despite perturbations

3.1 The Geometry of Biological Order

A system's tendency toward order depends on its geometry, which is determined by its information density. Since information (embodied in the system's material structure) is recovered from dissipated energy, a crucial relationship emerges both energy dissipation and information correlate more strongly with surface area than with volume. (Bigatti, D. & Susskind, L., 2000)

This reflects the **holographic principle**: within any bounded spatial region, entropy (a measure of system information) scales with surface area rather than volume.

3.2 The Information Density Limit

As organisms dissipate energy, they accumulate information. However, their finite surface area imposes a fundamental limit on recoverable information density. This creates an intimate energy-information relationship where: (Bekenstein, J., 2003)

1) Growth occurs through information accumulation from dissipated energy

2) Growth simultaneously reduces surface-to-volume ratio

Consequently:

- Energy dissipation capacity declines with size (being surface-dependent)
- Information density approaches its theoretical maximum (the Bekenstein bound)

This limit, formally described by Jacob Bekenstein, defines the conditions where a spatial region gains a new degree of freedom upon reaching maximum information density - effectively causing an n-dimensional space to exhibit (n+1)-dimensional properties at the boundary. (Bekenstein, J., 2003)

3.3 Critical Distinctions

Proper understanding requires careful differentiation between:

- Absolute size vs. dimensional scaling
- Information content vs. information density
- Surface-mediated processes vs. volumetric constraints

4. Size and Dimension in Biological Systems

4.1 Fundamental Definitions

- **Size**: The spatial extension of a material object, quantifiable as length (1D), area (2D), or volume (3D). Size represents magnitude without inherent reference to dimensionality.
- **Dimension**: The number of linearly independent basis vectors in a given space. Exemplified by:
  - Line (1D)
  - Plane (2D)
  - Volume (3D)

4.2 Key Distinction

Two circles differing in area possess distinct sizes but identical dimensionality (both being 2D planar figures). This separation becomes evident through surface curvature:

Illustrative Example:

A triangle drawn on:

1) A deflated balloon (2D planar surface)
2) The same balloon when inflated (3D curved surface)

While the triangle's size (edge lengths) remains constant, its embedding dimension increases from 2 to 3 through surface curvature.

4.3 Biological Implications

When an organism survives its growth phase boundary:

1) It reaches its **information density limit** (Bekenstein bound)
2) Its parameter space maintains size but gains dimensionality (+1 degree of freedom)
3) Energy-to-information recovery continues in this curved geometry

4.4 The Aging Mechanism

This dimensional transition induces:

- Progressive failure of variables to return to baseline values (geometric phase shift) (Barragán, J. & Sánchez, S., 2023)
- Declining system efficiency
- Emergence of aging as a geometric phenomenon

5. The Geometric Foundations of Biological Shape

The seminal question "Why is animal size so important?" posed by Knut Schmidt-Nielsen revolutionized biological thinking. The Norwegian naturalist's pioneering work established the fundamental relationship between organismal size and energy dissipation – perhaps his most enduring scientific legacy. (Schmidt-Nielsen, Knut, 1984)

5.1 Shifting the Paradigm to Shape

While understanding the determinants of biological morphology remains crucial, our focus centers on the tripartite relationship between *shape*, *size*, and *dimension*. Consider human embryogenesis: (Barragán, J. & Sánchez, S. 2023; Kaneko, K.J., 2016; Watanabe, T., Biggins, J.S., Tannan, N.B. & Srinivas, S., 2014)

1) **Week 1**: Spherical zygote (isotropic geometry)
2) **Week 2**: Bilaminar disc (planar topology)
3) **Week 4**: Cylindrical structure (neutral curvature)

This developmental trajectory - conserved across multicellular organisms — reveals a profound geometric principle: *Order emerges when system variables occupy spaces with*: (Solis Gamboa, D.A., 2010)

- Positive curvature (sphere)
- Null curvature (plane)

- Neutral curvature (cylinder)

5.2 Post-Growth Phase Transition

Upon reaching puberty's information density limit:

- Size stabilization occurs
- Dimensionality increases (+1 degree of freedom)
- Geometric phase shift manifests

Though macroscopic morphology remains apparently cylindrical, the system now operates in higher-dimensional space. (Jorge Barragán & Sebastián Sánchez, 2023)

This transition becomes detectable only through:

1) Aging-related variable drift
2) Progressive loss of homeostatic precision

The curvature of this emergent dimension presents detection difficulties analogous to temporal dimensions in spacetime - neither more nor less tractable than relativistic geometry. The aging process itself becomes the observable signature of this dimensional transformation. (Weisstein, E.W., 2024; Einstein, A., 1916; Thaheld, F.H., 2005)

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Historical Concepts of Disconnection in the Pioneering Works of Bleuler and Wernicke

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Abstract

The importance of terminology is a long-debated issue in psychiatry, and the original use of a term might give important clues to the following diagnostic notions and classifications. One of the pioneers of the modern psychiatry, Bleuler, has coined the term schizophrenia while in his extensive writings he speaks about several forms of splitting of the mental processes. Another major contemporary author to him, Wernicke, adopted another position but the final concept (sejunction) was another intriguing form of talking about disrupted mental processes. While considering the historical character of the work of both these renowned physicians, we have tried to lend some more perspective into the initial concepts of disconnection, as the modern neuroscience is calling it.

Keywords: schizophrenia, splitting, Bleuler, Wernicke, sejunction hypothesis

1. Introduction

The notion of disconnectivity and its subtleties must have a longer history from what demonstrated quite recently with tractography, magnetic resonance imaging and other sophisticated techniques. In fact, the intuition of pioneers of the modern psychiatry has coined some predecessors of the terminology.

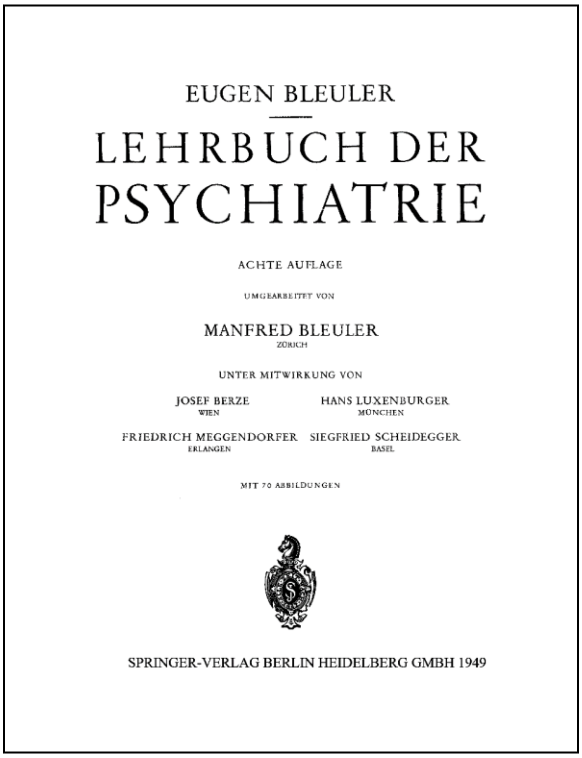
The elaborated notion that schizophrenia might be a disconnection syndrome has several sources. Not clearly an anatomical injury, but rather a condition with abnormal interaction or integration between cortical areas: this seems to be the theoretical hallmark of a diversity of studies (Friston, 1995; Schmitt, 2011).

The coiner of the term schizophrenia had already meant what the core of the issue was from Greek *skhizein*, 'to split', also been anglicized as σχίσμα (*schism*). While using this famous term for a diagnosis as old as humanity itself, Bleuler in 1911 probably forecasted the immensity of research would follow with regard to the fact that the patient with this occurrence has a splitting, but was not the same notion as that of multiple personality (Kyziridis, 2005).

In 1911, Bleuler noted: "*I call dementia praecox schizophrenia because, as I hope to show, the splitting of the different psychic functions is one of its most important features*" (Ashok, 2012). It was just the budding of a very large conceptual work, as we will try to show below, and as different sources point it out (Ljubičić, 2007).

2. Between 'Spaltung', 'Splitting', and Other Terms

Splitting of psychic functions was designated as '*Spaltung*' right from the initial work of Bleuler. In his *Textbook of Psychiatry* (Lehrbuch der Psychiatrie), he mentioned the term several times, and with different accessions (Bleuler, 1930; Bleuler, 1949).


EUGEN BLEULER

LEHRBUCH DER PSYCHIATRIE

ACHTE AUFLAGE

UMGEARBEITET VON

MANFRED BLEULER
ZÜRICH

UNTER MITWIRKUNG VON

JOSEF BERZE
WIEN

HANS LUXENBURGER
MÜNCHEN

FRIEDRICH MEGGENDORFER
ERLANGEN

SIEGFRIED SCHEIDEGGER
BASEL

MIT 70 ABBILDUNGEN

SPRINGER-VERLAG BERLIN HEIDELBERG GMBH 1949

Figure 1. Frontispiece of Bleuler's *Textbook of Psychiatry* in German, 8th edition

Here below in the Table 1, we summarize the original wording of the '*Spaltung*' as a concept, the respective chapter, and the English translation (Bleuler, 1949).

Table 1. 'Spaltung' and its subtleties in the Bleuler's *Textbook of Psychiatry*, 8th edition

Chapter	Page	Original in German	Translation
Die Störungen der Affektivität (Affective disorders)	**64**	Der Einfluß der Affekte auf die Assoziationen schafft Wahnideen, systematische **Zerspaltungen** der Persönlichkeit, hysteriforme Dämmerzustände […]	The influence of affects over the associations will produce delusions, systematic fragmentation of the personality, hysterical trances […]
Störungen der zentrifugalen Funktionen (Disorders of the centrifugal functions)	**80**	Bei den meisten Persönlichkeitsstörungen handelt es sich um **Spaltungen** nach affektiven Bedürfnissen […]	The majority of personality disorders has to do with the splitting of the affective needs […]
Vergiftungen (Intoxications)	**243**	Mit allerlei schizophrenen Zuständen haben die akuten Episoden des Cocainismus recht viel Ahnlichkeit: Körperhalluzinationen,	As with all kind of schizophrenic conditions, the episodes of acute cocainism are also very similar: somatic

		Stimmen, Wahnideen, **Spaltung** des Bewußtseins […]	hallucinations, voices, delusions, splitting of the consciousness […]
Vergiftungen (Intoxications)	**243**	Man kann aber mit den Cocaindeliranten leicht einen guten affektiven Kontakt bekommen; in der **Bewußtseinsspaltung** fällt unter anderem auf, daß in der Regel eine bestimmte Art Krankheitseinsicht vorhanden ist […]	However, during the cocaine delirium you may easily get an affective contact; for the splitting of the consciousness is of a certain form that as a rule there is still a certain disease insight […]
Die Schizophrenien (Dementia praecox) Schizophrenias (Dementia praecox)	**277**	[…] Bezeichnung "Dementia praecox" als unhaltbar. E. Bleuler hat sie 1911 durch diejenige der *Schizophrenie* ersetzt, weil ihm die elementarsten Störungen in einer mangelhaften Einheit, in einer Zersplitterung und **Aufspaltung** des Denkens […]	[…] The denomination "Dementia praecox" as not sound. E. Bleuler in 1911 replaced it with *Schizophrenia*, as the elementary disorders are due to a scarce unity, and a split as well as breakdown of the thought […]
Die Schizophrenien (Dementia praecox) Schizophrenias (Dementia praecox)	**283**	[…] habe ich einmal eine solche **Spaltung** des Gefühlsausdruckes gesehen.	[…] I have never seen such a split in the emotional expression.
Die Schizophrenien (Dementia praecox) Schizophrenias (Dementia praecox)	**284**	Die schizophrene **Funktionsspaltung** macht es möglich, daß Gegensätze, die sich sonst ausschließen, nebeneinander in der Psyche existieren.	The functional splitting in schizophrenia makes it possible, that opposites, which would exclude each other, might still co-exist in the psyche.
Die Schizophrenien (Dementia praecox) Schizophrenias (Dementia praecox)	**285**	Am bezeichnendsten aber sind die inneren **Spaltungen** des Willens. Die Kranken wollen etwas und zugleich das Gegenteil […]	Most impressive is the internal splitting of the will. The patients want something, and at the same time, the opposite […]
Die Schizophrenien (Dementia praecox) Schizophrenias (Dementia praecox)	**288**	[…] dieselbe schizophrene Denkstörung, wenn man \Ton Mangel an Ziel\Torstellung, Überwiegen der Nebenassoziationen, mangelhaftem Zusammenhang oder **Spaltung** des Denkens […]	[…] the same thought disorder in schizophrenia, if we consider lack of goal setting, predominance of loose associations, poor coherence or splitting of thought […]
Die Schizophrenien (Dementia praecox) Schizophrenias (Dementia praecox)	**302**	Das Gedanken-Hören an Stelle des automatischen Denkens bedeutet einen weiteren Schritt in der Entfremdung und **Abspaltung** von gewissen Vorstellungen vom Ich.	Hearing thoughts instead of automatic thinking represents a further step in the alienation and detachment of certain ideas from the self.

As one might conclude easily from this summarizing table, the *Spaltung* (splitting) in its initial form was further elaborated from Bleuler into disambiguations as *Zerspaltung, Aufspaltung* and *Abspaltung*, respectively fragmentation, breakdown, and detachment. Obviously, it might be difficult to find the right translation to very similar words that Bleuler, at that time, has been using purposefully to denote differences in clinical symptomatology. Furthermore, authors have synonymized Bleuler's detachment with the autistic withdrawal (Raja, 2010).

With very marginal and subtle differences, however these notions (*Spaltung, Zerspaltung, Aufspaltung* and *Abspaltung;* namely *splitting, fragmentation, breakdown* and *detachment*) were not casually used and should have underscored different grades of disconnectivity. Of course, if we stand firm to the synonymy of splitting with a disconnection occurrence.

3. Discussion

Historical sources have consistently focused on the disintegration of the psyche as a functional disorder, not an anatomical one: this is as well the pioneering concept of Bleuler' *schisis* (Friston, 2016).

From a different standing point, Wernicke, while representing primarily the neurological school he founded and that his followers were righteously proud of, come close to the notion. *Sejunction* hypothesis and other details from his work might be well only of historical value, but this clearly shed light on very debated and well-advanced neuropsychiatric opinions (Ungvari, 1993).

It is worth mentioning that Bleuler debated relentlessly and his psychiatric position was in fact, substantially different from the Wernicke-Kleist school. Wernicke was neurologically oriented, to him "*sejunction meant disruption in the interconnections between different neural systems, thus giving rise to loss of function, hyperfunction, and parafunction*" (Ungvari, 1993).

Wernicke holds as well the laurels of the association theory, with the idea that "*idea that pathological disorders of the central nervous system (which included mental illness) were caused by interruptions in the continuity of the association pathways*" (Lanczik, 1991). Moreover, "the location of the 'sejunction process' (***Sejunctionsvorganges***) would determine the *'Reizsymptom'* (irritation symptoms)" (Lanczik, 1991).

Might be actually difficult to draft conceptual contradictions between the *Spaltung* of Bleuler and the *Sejunction* of Wernicke; however, these two authors seem not to agree with each other. Maybe the professional-positional differences clearly related to the professional background of these two giants of the neuropsychiatry of XIX century:

> *The neuropathological explanation of psychotic symptoms Bleuler deemed to be a fruitless endeavor. In contrast, he favored psychological explanations, e.g. of psychomotor disturbances in schizophrenia. In this respect Bleuler drew on his large clinical experience, but he certainly also was influenced by psychoanalysis* (Pillmann, 2004).

Possibly the sejunction of Wernicke was as well a precursor to the actual notion of disconnectivity; if Wernicke based his findings on neuropathology, Bleuler was clearly a highly devoted psychiatrist; thus, the data and concepts he coined were fruit of clinical examination, accuracy and acumen. Both did not live in the era of sophisticated brain imaging, and could not find radiological proof of what their intuition suggested.

Even more, the selection of terms from Bleuler (Spaltung, Zerspaltung, Aufspaltung, and Abspaltung) was not merely a wording choice. To him for example, *Zerspaltung* was a systematic fragmentation of personality: thus, probably, a more serious condition leading to a poor prognosis, and a more severe clinical picture. The same must be valid for the other terms, which are not pure synonyms, as could be erroneously considered in a simplified form.

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Synergistic Regulation of LDL Receptor Expression by PCSK9 Inhibitors and Statins: A Molecular Review

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Abstract

Low-density lipoprotein receptor (LDLR) is a central mediator of plasma LDL-cholesterol (LDL-C) clearance. Statins enhance LDLR expression through SREBP2-mediated transcription but also upregulate proprotein convertase subtilisin/kexin type 9 (PCSK9), which targets LDLR for degradation. This feedback mechanism attenuates the cholesterol-lowering efficacy of statins. PCSK9 inhibitors, including monoclonal antibodies and siRNA-based therapies, prevent LDLR degradation and potentiate statin-induced LDLR upregulation. This review summarizes the molecular interplay between statins and PCSK9, explores their dual-axis impact on LDLR density, and evaluates outcome data from major clinical trials such as ODYSSEY, FOURIER, and ORION. The evidence supports a synergistic model wherein co-therapy enables profound LDL-C reduction and cardiovascular risk attenuation. Emerging approaches—including gene editing, antisense oligonucleotides, and integrative lipidomic-guided therapy—suggest a future of increasingly precise and durable modulation of LDLR activity.

Keywords: LDL receptor, PCSK9, statins, cholesterol metabolism, SREBP2, monoclonal antibody, Inclisiran, LDL-C, cardiovascular prevention, gene regulation

1. The Central Role of LDL Receptor in Cholesterol Homeostasis

The low-density lipoprotein receptor (LDLR) is a transmembrane glycoprotein predominantly expressed in hepatocytes, where it mediates the endocytosis of circulating low-density lipoprotein cholesterol (LDL-C), the primary carrier of plasma cholesterol. Through specific recognition and binding of apolipoprotein B-100 (ApoB-100) on the surface of LDL particles, LDLR initiates clathrin-dependent internalization of LDL into liver cells, followed by lysosomal degradation of the lipoprotein cargo and recycling of the receptor to the plasma membrane. This pathway accounts for the clearance of approximately 70% of circulating LDL-C under physiological conditions, underscoring LDLR's indispensable role in systemic cholesterol regulation.

The expression and activity of LDLR are tightly controlled by intracellular cholesterol levels via the sterol regulatory element-binding protein 2 (SREBP2) pathway. When intracellular cholesterol levels fall, SREBP2 is cleaved and translocated into the nucleus, where it binds to the sterol regulatory elements (SREs) within the LDLR gene promoter to enhance transcription. This feedback loop ensures that hepatocytes upregulate LDLR synthesis in response to cholesterol demand, thereby lowering plasma LDL-C.

LDLR activity also intersects with broader metabolic and inflammatory signals. For instance, insulin and thyroid hormone stimulate LDLR expression, whereas inflammatory cytokines such as tumor necrosis factor-alpha (TNF-α) and interleukin-1β may downregulate its transcriptional or post-translational stability. Moreover, LDLR-deficient mouse models and familial hypercholesterolemia (FH) patients with LDLR gene mutations exhibit markedly elevated LDL-C levels and accelerated atherosclerosis, providing robust in vivo and clinical evidence of LDLR's pivotal role in lipid metabolism.

Despite its efficiency, LDLR's impact on cholesterol clearance is modulated by its post-translational regulators, most notably proprotein convertase subtilisin/kexin type 9 (PCSK9), which promotes LDLR lysosomal degradation. This negative regulation limits LDLR availability at the cell surface and, by extension, the maximal achievable LDL-C clearance, even under conditions of high transcriptional activation (e.g., with statin therapy). This molecular bottleneck has made LDLR a strategic pharmacologic node, with modern lipid-lowering therapies aiming to preserve or enhance LDLR expression and recycling.

Thus, LDLR functions as a molecular gatekeeper in cholesterol homeostasis—its abundance, stability, and activity directly shaping plasma LDL-C concentrations. Interventions that increase LDLR expression or prevent its degradation form the cornerstone of contemporary lipid management strategies, particularly for high-risk cardiovascular patients.

2. Statin-Induced Upregulation of LDLR: Mechanisms and Limitations

2.1 HMG-CoA Reductase Inhibition and SREBP2 Activation

Statins (e.g., atorvastatin, rosuvastatin, simvastatin) exert their lipid-lowering effects by inhibiting 3-hydroxy-3-methylglutaryl-CoA reductase (HMGCR), the key rate-limiting enzyme in the mevalonate pathway of cholesterol biosynthesis. This blockade results in a decrease in intracellular cholesterol pools in hepatocytes, triggering a homeostatic transcriptional response to replenish cholesterol levels.

The primary effector of this response is sterol regulatory element-binding protein 2 (SREBP2), a membrane-bound transcription factor that remains inactive in the endoplasmic reticulum under cholesterol-replete conditions. Upon depletion of intracellular sterols, SREBP2 is cleaved in the Golgi by S1P and S2P proteases, releasing its N-terminal active domain. This domain translocates into the nucleus and binds sterol response elements (SREs) in the promoters of target genes—including LDLR, HMGCR, and PCSK9.

The activation of SREBP2 leads to a robust transcriptional upregulation of LDLR, enhancing the hepatic uptake of circulating LDL particles. As LDLR density on the hepatocyte surface increases, so does the endocytosis and clearance of LDL-C from plasma. Quantitatively, this pathway explains why moderate-to-high intensity statin therapy can reduce plasma LDL-C levels by 30–55%, depending on dosage, statin potency, and baseline cholesterol status.

Statins also induce LDLR mRNA stability and facilitate receptor recycling from endosomes, further enhancing LDLR function beyond transcriptional control. These mechanisms are dose-dependent but also subject to diminishing returns, as seen in the "rule of 6" in statin pharmacodynamics: every doubling of the statin dose results in only ~6% further LDL-C reduction. This saturation effect is in part due to counterregulatory pathways, including the one involving PCSK9.

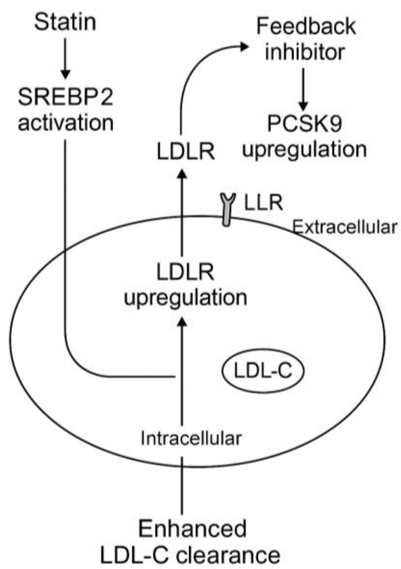

Figure 1. SREBP2-Mediated Dual Regulation of LDL Receptor and PCSK9 Under Statin Therapy

2.2 Feedback Elevation of PCSK9 Expression

While statins boost LDLR expression, they concurrently activate PCSK9, a secreted serine protease that binds LDLR and targets it for lysosomal degradation. This dual regulation arises because PCSK9 is also an SREBP2-responsive gene. The same transcription factor that increases LDLR transcription also elevates PCSK9 mRNA and protein levels, creating a molecular tug-of-war between LDLR synthesis and degradation.

Clinically, this manifests as partial attenuation of statin efficacy. After several weeks of statin use, serum PCSK9 concentrations rise significantly—by 35% to 70% depending on the statin type and dose (Dubuc et al., 2004; Mayne et al., 2008). This increase correlates with a plateau in LDL-C reduction, even when statin doses are intensified, as more newly synthesized LDLR are degraded before reaching or remaining on the cell surface.

Notably, individuals with higher baseline PCSK9 levels or genetic variants that lead to gain-of-function mutations (e.g., p.S127R or p.D374Y) show blunted responses to statins, further validating PCSK9 as a dominant regulator of statin efficacy. Conversely, individuals with PCSK9 loss-of-function mutations (e.g., p.Y142X or p.R46L) experience greater LDL-C reduction with statins and lower lifelong cardiovascular risk, underscoring PCSK9's pivotal role in statin pharmacogenetics.

Additionally, PCSK9-mediated degradation of LDLR becomes more problematic in patients with familial hypercholesterolemia (FH), where residual LDLR activity is already compromised. In such contexts, statins alone are often insufficient, and adjunctive PCSK9-targeted therapies become necessary to achieve guideline-recommended LDL-C targets.

This feedback effect has shifted the clinical strategy from "statin maximization" to statin–PCSK9 inhibitor co-therapy, especially in secondary prevention populations or those with statin intolerance or resistance. By suppressing PCSK9-induced LDLR degradation, clinicians can unmask the full potential of statin-induced LDLR transcription, leading to additive or even synergistic effects on LDL-C lowering.

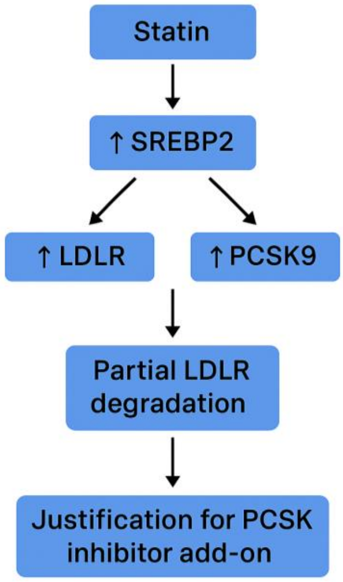

Figure 2. Dual Regulatory Effects of Statins on LDL Receptor and PCSK9 Expression

3. PCSK9 Biology and Its Role in LDLR Degradation

3.1 PCSK9–LDLR Binding and Endosomal Targeting

PCSK9 is synthesized as a 692-amino acid zymogen predominantly in the liver and undergoes autocatalytic cleavage in the endoplasmic reticulum (ER) to generate a mature, secreted form. Although classified as a serine protease, PCSK9's physiological activity does not rely on catalytic cleavage of other proteins. Instead, it functions primarily through protein–protein interaction, particularly with the epidermal growth factor-like repeat A (EGF-A) domain of LDLR.

Upon secretion, circulating PCSK9 binds to surface LDLRs in a calcium-dependent manner, forming a stable complex. When this complex is endocytosed via clathrin-coated pits, the acidic environment of the endosome fails to dissociate PCSK9 from LDLR, unlike LDL particles that are readily released at low pH. This persistent interaction redirects LDLR from its usual recycling route to lysosomal degradation, decreasing LDLR half-life on the cell surface and thereby limiting hepatic clearance of LDL-C.

Crystallographic studies have shown that PCSK9 binding alters LDLR's conformation, rendering it unrecognizable by sorting nexin proteins involved in recycling. This explains why even modest elevations in PCSK9 levels result in substantial reductions in functional LDLR density. Notably, unlike LDL itself, PCSK9 binds to both occupied and unoccupied LDLRs, amplifying its regulatory potential even in the absence of ligand.

Moreover, hepatocyte-specific knockout of PCSK9 in mice leads to marked increases in hepatic LDLR abundance and a ~80% reduction in plasma LDL-C, confirming the suppressive effect of endogenous PCSK9 in vivo. These findings underscore the post-translational, receptor-limiting role of PCSK9 and justify its inhibition as a therapeutic approach.

3.2 Regulation by Nutrient and Hormonal Signals

While SREBP2 is the primary transcriptional driver of PCSK9, its expression is fine-tuned by a complex network of systemic and intracellular signals, reflecting the need to balance cholesterol clearance with cell membrane integrity and metabolic demand.

Key modulators of PCSK9 transcription and secretion include:

- **Insulin and feeding state:** PCSK9 mRNA is upregulated in the fed state via insulin-stimulated HNF1α activity. This explains why patients with type 2 diabetes or metabolic syndrome often present with elevated PCSK9 levels, independent of cholesterol status.
- **Fasting and PPAR-α activation:** Nutritional deprivation or fibrate-induced PPAR-α activation suppresses PCSK9 transcription, possibly via hepatic lipid oxidation pathways.
- **Inflammatory cytokines:** IL-6 and TNF-α promote PCSK9 expression via the STAT3 and NF-κB pathways, respectively, linking inflammation to impaired cholesterol clearance. This is particularly relevant in chronic diseases such as rheumatoid arthritis or systemic lupus erythematosus, where LDL-C may paradoxically remain normal despite high cardiovascular risk—partly due to PCSK9-mediated LDLR suppression.
- **Estrogen and thyroid hormone:** Both have been shown to reduce PCSK9 levels, potentially contributing to sex- and thyroid-related lipid variations.
- **Circadian rhythm:** PCSK9 exhibits **diurnal variation**, peaking in the early morning and declining in the evening. This suggests potential for time-optimized dosing of PCSK9-targeted therapies to maximize efficacy.

These factors position PCSK9 as a metabolic integrator, responsive not only to sterol status but also to hormonal, inflammatory, and temporal signals. As such, therapeutic modulation of PCSK9 must consider both direct blockade and contextual modulation through upstream pathways.

3.3 Genetic Variants and Clinical Phenotypes

The discovery of PCSK9's role in human lipid physiology was propelled by population-based genetic studies. Initial clues came from families with autosomal dominant hypercholesterolemia (ADH) in whom no mutations in LDLR or ApoB were found. Sequencing revealed gain-of-function (GOF) mutations in PCSK9, such as D374Y, which increased its binding affinity to LDLR and promoted aggressive receptor degradation.

Patients carrying GOF mutations exhibit markedly elevated LDL-C (>250 mg/dL) and increased incidence of early-onset coronary artery disease. These findings validated PCSK9 not only as a regulator but also as a driver of atherogenesis when dysregulated.

Conversely, loss-of-function (LOF) mutations such as Y142X and R46L are associated with lifelong reductions in LDL-C of 15–40 mg/dL and a >80% reduction in coronary heart disease risk, as shown in seminal studies by Cohen et al. (2006) and further supported by Mendelian randomization analyses.

What makes these findings particularly impactful is the absence of major adverse effects in LOF carriers. Despite very low LDL-C levels from early life, these individuals show no increased risk of liver dysfunction, hormonal imbalance, or cognitive impairment. This long-term safety profile strongly supports the therapeutic inhibition of PCSK9, even in aggressive or lifelong formats (e.g., siRNA).

Notably, PCSK9 mutation frequency varies by ethnicity. For instance, R46L is more prevalent in European populations, while Y142X is relatively enriched in African-derived populations. This has implications for personalized lipid-lowering strategies and highlights the value of genetic screening in statin-resistant hypercholesterolemia.

4. Mechanisms of PCSK9 Inhibition: From Antibodies to siRNA

4.1 Monoclonal Antibodies: Alirocumab and Evolocumab

Monoclonal antibodies (mAbs) targeting PCSK9 were the first therapeutic agents approved to inhibit its function. Alirocumab and Evolocumab are fully human IgG-based antibodies that bind circulating PCSK9 with high specificity and affinity, preventing its interaction with the LDL receptor. This blockade preserves LDLR recycling and dramatically enhances hepatic LDL-C clearance.

These agents function extracellularly, neutralizing PCSK9 before it binds LDLR. Administered subcutaneously, both drugs exhibit favorable pharmacokinetics, with half-lives of approximately 11–20 days, allowing biweekly or monthly dosing. Clinical trials, including ODYSSEY OUTCOMES and FOURIER, have demonstrated that these antibodies lower LDL-C by ~60% on top of statin therapy, and reduce cardiovascular events by 15–20% in high-risk populations (Sabatine et al., 2017; Schwartz et al., 2018).

Mechanistically, the mAbs do not reduce PCSK9 production; instead, they increase its clearance via immune complex formation, which is then degraded in the liver. This explains the sustained reduction in free PCSK9 and corresponding rise in functional LDLR density.

An advantage of monoclonal antibodies is immediate onset of action and well-characterized dose–response relationships. However, they are relatively expensive and require chronic administration, with long-term adherence being a concern in non-injectable-tolerant populations.

4.2 siRNA Agents (Inclisiran) and Post-Transcriptional Control

A newer modality for PCSK9 inhibition involves small interfering RNA (siRNA) technology, exemplified by Inclisiran—a GalNAc-conjugated, double-stranded siRNA that targets PCSK9 mRNA in hepatocytes. Administered subcutaneously, Inclisiran utilizes receptor-mediated endocytosis via the asialoglycoprotein receptor (ASGPR), achieving hepatocyte-specific gene silencing.

Once inside the cell, the antisense strand of the siRNA duplex is loaded onto the RNA-induced silencing complex (RISC), which binds and cleaves PCSK9 mRNA, leading to reduced synthesis of both intracellular and secreted PCSK9. This reduction translates into increased LDLR availability, with LDL-C lowering comparable to mAb therapy (~50–55% reduction).

One of Inclisiran's most notable features is its long duration of action. Because it affects intracellular synthesis rather than extracellular neutralization, its pharmacodynamic effect lasts much longer—requiring dosing only twice per year after the initial two doses at day 0 and 90. This improves adherence and lowers treatment burden.

Unlike antibodies, Inclisiran achieves lower steady-state PCSK9 levels rather than transient spikes and troughs. However, it has a slower onset of action, taking several weeks to reach maximal LDL-C reduction, which may be a consideration in acute settings or post-ACS scenarios.

Overall, siRNA therapy represents a novel mechanistic paradigm: not blocking PCSK9 after it's made, but preventing its synthesis altogether, offering durable, low-frequency intervention with a favorable safety profile.

4.3 Comparative Pharmacokinetics and Tissue Penetration

From a pharmacokinetic perspective, monoclonal antibodies and siRNA differ markedly:

Table 1.

Parameter	**Monoclonal Antibodies (Alirocumab/Evolocumab)**	**siRNA (Inclisiran)**
Mechanism of Action	Extracellular binding of PCSK9	Intracellular mRNA degradation
Time to Peak Effect	Days	Weeks (up to 30–45 days)

Dosing Frequency	Every 2 or 4 weeks	Twice per year (after loading phase)
Site of Action	Circulating PCSK9	Hepatocyte PCSK9 production
Clearance Mechanism	RES and hepatic catabolism of immune complexes	RNAi degradation and hepatic RISC

From a tissue-targeting standpoint, both approaches are liver-focused, as hepatic PCSK9 contributes the vast majority of systemic PCSK9 levels. However, Inclisiran's GalNAc-targeting ensures near-exclusive hepatocyte uptake, minimizing systemic exposure and enhancing safety.

Emerging modalities—such as vaccines, antisense oligonucleotides (ASOs), and gene-editing approaches (e.g., CRISPR-Cas9-based PCSK9 knockdown)—are under investigation but remain in early clinical stages.

PCSK9 inhibition now exists across multiple mechanistic layers: from extracellular protein neutralization to intracellular gene silencing. Each modality has its own kinetic profile, patient suitability, and clinical niche. Their development reflects the evolution of LDLR-based therapy from statin-anchored strategies toward precision modulation of receptor turnover, establishing a foundation for the synergistic co-therapies explored in subsequent sections.

5. Synergistic Effects of Statins and PCSK9 Inhibitors on LDLR Density

5.1 Statin-Induced PCSK9 Upregulation and the Rationale for Co-Therapy

While statins remain the cornerstone of lipid-lowering therapy, their mechanism of action paradoxically includes the upregulation of PCSK9, which can attenuate their full therapeutic potential. As discussed previously, statins activate the SREBP2 transcriptional axis, which simultaneously induces both LDLR and PCSK9 gene expression. The result is a compensatory feedback loop, whereby newly synthesized LDL receptors are targeted for degradation by rising PCSK9 levels.

This paradox has been confirmed in both mechanistic studies and clinical data. Statin monotherapy often reaches a plateau in LDL-C reduction, particularly at high doses, with only marginal gains despite increased LDLR transcription. This phenomenon—sometimes termed the "PCSK9 ceiling effect"—has been shown to limit the maximal LDL-C lowering to approximately 50–55% even with potent statins like rosuvastatin at 40 mg daily.

The rationale for co-therapy thus becomes molecularly evident: while statins induce LDLR transcription, PCSK9 inhibitors preserve LDLR from degradation. This pairing allows for a two-pronged enhancement of receptor availability—increasing production while minimizing loss.

By inhibiting PCSK9 (via monoclonal antibody or siRNA), this compensatory degradation pathway is suppressed, allowing the full expression of statin-induced LDLR synthesis to manifest as increased LDL-C clearance. Mechanistically, statins "step on the gas" by producing more receptors, while PCSK9 inhibitors "release the brakes" by preventing their removal. The result is a synergistic, rather than merely additive, increase in LDLR density at the hepatocyte surface.

5.2 Dual Modulation of LDLR Recycling and Degradation Pathways

From a systems biology perspective, LDLR availability on the hepatocyte membrane is the net result of:

- Rate of synthesis (transcriptional and translational)
- Intracellular trafficking efficiency
- Endosomal recycling rate
- Rate of lysosomal degradation (primarily PCSK9-mediated)

Statins act primarily at the first stage, increasing transcription and translation of LDLR through SREBP2 activation. PCSK9 inhibitors act at the fourth stage, decreasing degradation of surface-expressed LDLR. Together, they enable:

- Increased receptor abundance
- Prolonged receptor half-life
- Sustained LDL-C clearance capacity

Recent in vitro studies using hepatocyte cell lines (e.g., HepG2) show that statin + PCSK9 inhibitor combination increases LDLR surface expression by 2.5–3.0 fold compared to statin alone. In vivo, these changes translate to ~60–70% LDL-C reduction, compared to ~45–55% with statins alone, or ~50–60% with PCSK9 inhibitors as monotherapy.

This synergy is not merely pharmacodynamic but also pharmacoeconomic. By allowing for lower doses of each agent while achieving greater effect, combination therapy may reduce adverse event rates (e.g., statin-associated muscle symptoms) and improve patient tolerability and adherence.

Furthermore, the combination is particularly critical in patients with:

- Familial hypercholesterolemia (FH) – where residual LDLR activity is limited
- Statin intolerance – where LDLR induction is suboptimal
- High-risk secondary prevention – where LDL-C targets <55 mg/dL are often unattainable with statins alone

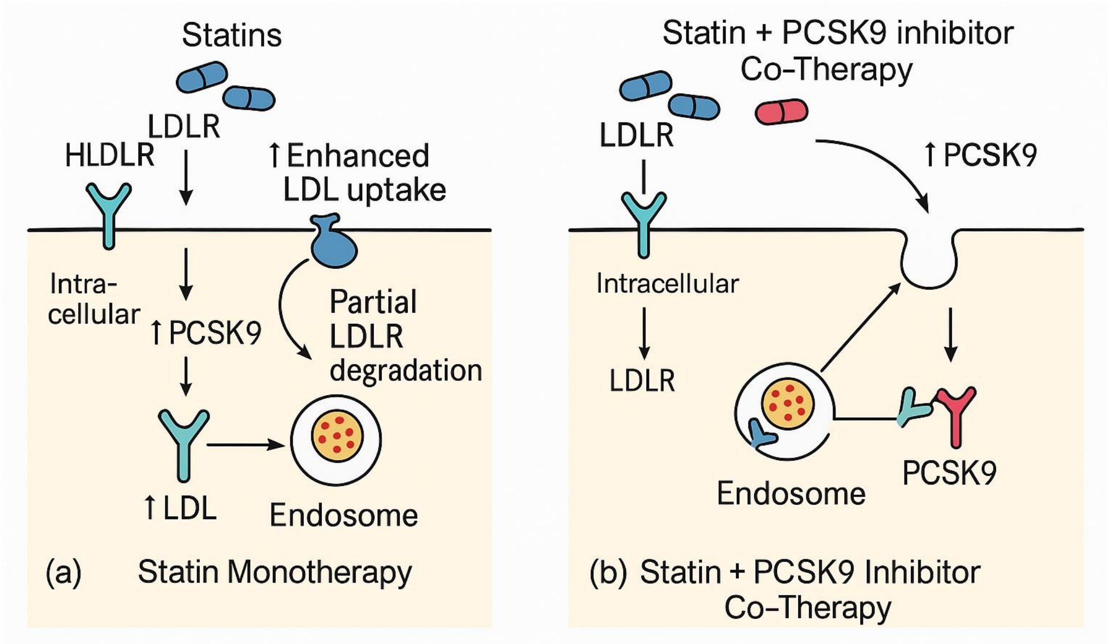

Figure 3. LDLR lifecycle under (a) statin monotherapy and (b) statin + PCSK9 inhibitor co-therapy

In summary, the dual modulation of LDLR production and protection forms the mechanistic basis for modern lipid-lowering synergy, enabling aggressive LDL-C targets to be met safely and effectively. The upcoming clinical section will explore how this molecular synergy translates into real-world cardiovascular outcome benefits.

6. Clinical Outcomes and Biomarker Evidence of Synergy

The molecular synergy between statins and PCSK9 inhibitors—via combined LDLR upregulation and protection—translates into substantial clinical benefits. A growing body of evidence from large-scale randomized controlled trials (RCTs) confirms that this co-therapy not only achieves deeper LDL-C reductions but also results in significant decreases in atherosclerotic cardiovascular disease (ASCVD) events, particularly in high-risk populations.

LDL-C Reduction and Atherosclerotic Cardiovascular Risk

In most trials, the addition of a PCSK9 inhibitor to statin therapy results in an additional 50–65% reduction in LDL-C levels, beyond what statins alone can achieve. This magnitude of LDL-C lowering has been consistently associated with proportional reductions in cardiovascular risk, consistent with the "lower is better" hypothesis established by meta-analyses of statin trials.

Moreover, combination therapy allows a substantial proportion of patients to reach very low LDL-C thresholds (e.g., <55 mg/dL or even <30 mg/dL) that are now endorsed by international guidelines (ESC/EAS, AHA/ACC) for patients at very high risk.

Data from ODYSSEY, FOURIER, and ORION

Trials

1) **ODYSSEY OUTCOMES (Alirocumab + Statin)**

Population: ~18,000 patients post-acute coronary syndrome (ACS)

- Result: Alirocumab added to high-intensity statins reduced LDL-C from a median of 92 to 53 mg/dL at 12 months
- CV Outcome: 15% relative risk reduction (RRR) in major adverse cardiovascular events (MACE) over 2.8 years
- Notably, in patients with baseline LDL-C >100 mg/dL, the risk reduction rose to 24%, demonstrating benefit stratified by baseline risk (Schwartz et al., 2018)

2) **FOURIER (Evolocumab + Statin)**

- Population: ~27,000 patients with stable ASCVD
- Result: Evolocumab reduced LDL-C by 59% (from 92 to 30 mg/dL), sustained over a median of 2.2 years
- CV Outcome: 15% RRR in MACE; 20% RRR in composite hard endpoints (CV death, MI, stroke) at 3 years
- Subgroup analyses showed greatest benefit in patients achieving LDL-C <25 mg/dL, with no increase in adverse events or neurocognitive decline (Sabatine et al., 2017)

3) **ORION-10 and ORION-11 (Inclisiran + Statin)**

- Population: >3,000 patients with ASCVD or risk equivalents
- Result: Twice-yearly Inclisiran led to 51% LDL-C reduction vs placebo when added to statins
- Long-term data from ORION-4 (expected 2026) will assess cardiovascular outcomes
- Early biomarker evidence shows sustained PCSK9 suppression and LDLR upregulation with excellent tolerability

Beyond LDL-C: Inflammatory and Plaque Stabilization Markers

While LDL-C remains the primary surrogate marker, exploratory analyses have revealed additional benefits of PCSK9 inhibition:

- **High-sensitivity C-reactive protein (hsCRP):** Although not significantly lowered by PCSK9 inhibitors, hsCRP baseline stratification suggests patients with elevated inflammation may derive greater absolute risk reduction, especially when combined with statin-driven anti-inflammatory effects.
- **Plaque regression:** Imaging studies (e.g., GLAGOV trial) demonstrated that Evolocumab + statin therapy led to significant coronary plaque volume regression, a rare outcome in lipid trials, aligning with real reductions in ischemic risk.
- **Lipoprotein(a) reduction:** PCSK9 inhibitors reduce Lp(a) levels by ~25%, offering added benefit in patients with genetically elevated Lp(a)—a population poorly addressed by statins.

Together, these findings affirm that PCSK9 inhibitors, when combined with statins, amplify lipid-lowering, improve vascular outcomes, and reduce event burden across a range of high-risk patient populations. Their favorable safety profiles, even at LDL-C levels once considered "too low," support aggressive lipid-lowering as a safe and effective long-term strategy.

The evidence base strongly validates the molecular logic: increasing LDLR expression (via statins) and preventing its degradation (via PCSK9 inhibition) results in sustained therapeutic efficacy with measurable benefit on clinical endpoints.

7. Future Therapeutic Strategies and Molecular Optimization

The success of PCSK9-targeted therapies and statin co-treatment has reinvigorated interest in refining lipid-lowering strategies at both the molecular and clinical levels. As the demand for durable, cost-effective, and individualized therapies increases, future approaches are likely to move beyond protein-level modulation toward gene-targeted, combinatorial, and adaptive strategies.

7.1 Next-Generation PCSK9 Modulators

One frontier in lipid regulation lies in gene editing technologies, especially CRISPR-Cas9–based systems designed to introduce permanent loss-of-function (LOF) mutations in PCSK9. In preclinical models, a single dose of lipid nanoparticle–delivered CRISPR constructs has resulted in >90% reduction in circulating PCSK9 and sustained LDL-C lowering for over 12 months. Human trials (e.g., Verve Therapeutics' VERVE-101) are underway to evaluate the long-term efficacy and safety of this approach in heterozygous familial hypercholesterolemia (HeFH) patients.

Other investigational approaches include:

- **Antisense oligonucleotides (ASOs):** Unlike siRNA, ASOs bind to mRNA in the nucleus and reduce PCSK9 production at the transcriptional level. Early-generation compounds (e.g., AZD8233) show potent LDL-C reduction with once-monthly dosing.
- **Vaccines targeting PCSK9 epitopes:** These aim to induce long-lasting humoral immunity against PCSK9, offering the potential for annual or semi-annual dosing at low cost—particularly attractive for resource-limited settings.
- **Allosteric inhibitors and oral small molecules:** Efforts are ongoing to discover orally bioavailable compounds that modulate PCSK9–LDLR interaction without requiring injectable administration.

7.2 Dual and Triple Pathway Modulation

Beyond PCSK9, future lipid-lowering therapies may benefit from multi-target strategies that simultaneously address different facets of atherogenic lipoprotein metabolism. Examples include:

- **Bempedoic acid** (an ACL inhibitor): When added to statin + PCSK9 therapy, this agent provides further LDL-C reduction and anti-inflammatory benefits (lowering hsCRP), without increasing muscle-related side effects.
- **ANGPTL3 inhibitors** (e.g., evinacumab): Targeting this hepatic protein reduces both LDL-C and triglycerides, making it useful in patients with mixed dyslipidemia or residual risk after LDL-C control.
- **Lp(a)-lowering therapies:** Novel RNA-based agents (e.g., pelacarsen) under development may complement PCSK9 inhibitors in genetically predisposed individuals.

By combining statins, PCSK9 suppression, and other lipid pathway interventions, patients could achieve comprehensive atheroprotection with improved tolerability and adherence profiles.

7.3 Precision Lipidology and Systems-Level Integration

The future of LDLR regulation will not only be molecular but data-driven. Multi-omic platforms—including lipidomics, transcriptomics, and pharmacogenomics—can help stratify patients based on:

- Genetic responsiveness to statins or PCSK9 inhibition
- Residual inflammatory or thrombotic risk
- Polygenic lipid risk scores and familial markers

These datasets will enable personalized lipid-lowering plans that match drug choice, intensity, and target levels to individual biology. Artificial intelligence and clinical decision support systems may further refine therapy timing and sequencing, reducing overtreatment and improving outcomes.

At the systems biology level, mathematical models are being developed to simulate LDLR turnover, PCSK9 kinetics, and lipid flux across organs. Such computational frameworks will inform not only dosing schedules but also predict response trajectories to dual or triple therapy regimens, adapting therapy to patient-specific parameters.

In conclusion, the synergistic regulation of LDLR by statins and PCSK9 inhibitors represents both a mechanistic triumph and a clinical milestone in cardiovascular prevention. Looking forward, innovation will rely on translating this synergy into simpler, longer-lasting, and more personalized therapies, grounded in molecular precision and supported by evolving bioengineering platforms. The LDL receptor, once merely a receptor, now stands at the center of a multi-dimensional therapeutic architecture that is reshaping the future of lipidology.

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Knowledge, Attitude, and Behavior of Pediatric Medical Surgical Nurses on Breastfeeding: Basis for Capacity Building Plan

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Abstract

Breastfeeding in pediatric medical-surgical units has been shown to significantly reduce the incidence of various complications in hospitalized neonates, including necrotizing enterocolitis, infections, retinopathy of prematurity, and chronic lung disease. Furthermore, breastfeeding lowers both the incidence and mortality rates associated with long-term neurological injuries, making it particularly crucial for vulnerable populations such as preterm infants. Pediatric medical-surgical nurses play an essential role as advocates and facilitators of breastfeeding practices for hospitalized newborns, directly influencing the quality of care provided in these settings. Despite the well-documented benefits of breastfeeding, the lack of standardized management guidelines for neonatal breastfeeding across hospitals has created challenges for nursing staff in both educating families and implementing consistent breastfeeding practices. These inconsistencies may hinder the overall effectiveness of breastfeeding initiatives in improving neonatal health outcomes. This study aimed to evaluate the knowledge, attitudes, and practices of pediatric medical-surgical nurses regarding neonatal breastfeeding. By utilizing comprehensive questionnaires, the study assessed the current level of understanding, attitudes toward breastfeeding, and practical application of breastfeeding practices among local pediatric medical-surgical nurses. The findings from this research provided valuable insights into the specific factors that influence nursing practices and guided the development of standardized protocols and training programs. These efforts were intended to improve the implementation of neonatal breastfeeding practices, thereby enhancing health outcomes for hospitalized infants.

Keywords: neonatal breastfeeding, pediatric medical-surgical nurses, Knowledge Attitudes and Practices (KAP), standardized management guidelines, hospitalized neonates

1. Introduction

Breastfeeding is universally recognized as the optimal feeding method by international health organizations and various health institutions. Breast milk not only contains rich nutrients that are easily absorbed by infants, but it also includes multiple immune-active components that enhance an infant's immunity, helping them resist infections and promote healthy development (Lokossou et al., 2022). Beyond its essential role in infancy, breastfeeding is increasingly recognized for its importance in pediatric medical-surgical (MS) cases, particularly in post-surgical recovery. Breastfeeding offers non-nutritional benefits such as reducing post-operative pain, accelerating healing, preventing infections, and improving long-term health outcomes for pediatric patients (Elgersma et al.; Søegaard et al., 2024).

In recent years, there has been a global effort to promote breastfeeding. However, according to data from the World Health Organization (WHO), the exclusive breastfeeding rate for infants under six months was only 43% in 2019, which is significantly below the ideal target (World Health Organization, 2019). In China, the "Outline for the Development of Chinese Children (2011-2020)" set a goal for the exclusive breastfeeding rate for infants aged 0-6 months to exceed 50% by 2020 (Yan et al., 2023). Despite these objectives, the promotion and implementation of breastfeeding remain insufficient in many areas, particularly among hospitalized pediatric patients (Moura et al., 2022; Mudau et al., 2023).

Recent statistics indicate that approximately 10-21.2% of newborns and infants require hospitalization due to complications such as congenital anomalies, infections, or surgical needs (China Information News, 2021). This statistic translates to a substantial number of pediatric patients in China requiring specialized care in medical-surgical departments. However, these hospitalized pediatric patients often face challenges including limited parental visitation, postoperative complications, and a lack of structured breastfeeding support systems within hospital environments (Verea-Nuñez et al., 2024; Wu et al., 2024).

Moreover, the management of breastfeeding for hospitalized newborns currently lacks standardized protocols, leading to inconsistencies in nursing practices. For instance, many hospitals do not have clear guidelines regarding the timing and frequency of breastfeeding, resulting in variations in decision-making among healthcare providers. Additionally, nurses may encounter difficulties in educating mothers, such as a lack of effective educational materials and practical opportunities to provide necessary support. Some hospitals may fail to establish breastfeeding support groups or consulting services, which can prevent mothers in need of assistance from receiving timely guidance. Furthermore, the pressures of the hospital environment may limit nurses' ability to provide individualized breastfeeding education, ultimately affecting mothers' confidence and willingness to breastfeed. These issues underscore the necessity for clearer management protocols to enhance the effectiveness of breastfeeding implementation for newborns.

While there is a considerable body of literature on breastfeeding practices in neonatal intensive care units (NICUs), research specifically focused on the knowledge, attitudes, and practices (KAP) of pediatric surgical nurses regarding breastfeeding remains limited (Srichalerm et al., 2024). The pediatric surgical patient population has unique needs that require specific breastfeeding approaches to accommodate surgical recovery and medical treatment plans. Thus, there is a pressing need to develop a capacity-building plan that equips pediatric surgical nurses with the essential knowledge, skills, and support necessary to effectively promote breastfeeding in complex medical and surgical environments.

Therefore, this study aimed to systematically investigate the KAP of pediatric surgical nurses regarding breastfeeding and to identify the correlations between their KAP and demographic characteristics (e.g., age, education level, work experience). Through this analysis, the study sought to provide a foundation for developing targeted training programs and policies that would enhance the practical implementation of breastfeeding in pediatric surgical wards, ultimately improving patient recovery outcomes. The research framework was based on a comprehensive analysis of nurses' KAP, considering the specific needs of pediatric surgical patients, with the ultimate goal of creating a capacity-building plan for pediatric surgical nurses to improve breastfeeding practices and outcomes.

2. Study Objectives

2.1 General Objective

To explore factors related to the knowledge, attitude, and practices of pediatric medical surgical nurses regarding breastfeeding, which will be a basis for a health promotion plan.

2.2 Specific Objectives

1) To describe the demographic characteristics of the respondents as to: a) age, b) sex, c)

highest educational attainment, d) position category, e) years of nursing experience, f) years of pediatric MS experience, g) number of capacity building activity within one year;

2) To describe the knowledge, attitude, and practices (KAP) of the respondents related to breastfeeding;

3) To compare the KAP of the respondents on breastfeeding when grouped according to demographic characteristics;

4) To determine the relationship between knowledge, attitude, and practices (KAP) of the respondents about breastfeeding;

5) Based from the findings, to create a Capacity Building Plan for Pediatric Medical-Surgical Nurses related to Breastfeeding.

3. Review of Related Literature

3.1 Demographic Characteristics of Pediatric Medical-Surgical Nurses

The demographic characteristics of healthcare providers play a pivotal role in shaping their approach to patient care, including breastfeeding support. In the context of pediatric medical-surgical (MS) nursing, factors such as age, gender, educational background, and years of experience directly influence how nurses perceive and promote breastfeeding practices.

Research indicates that nurses' age and professional experience can significantly affect their attitudes toward breastfeeding (Al-Sawalha et al., 2018). Younger nurses, or those new to pediatric MS care, may lack the deep-seated knowledge or confidence needed to support breastfeeding, compared to their more experienced counterparts. Conversely, older nurses may be more resistant to adopting new breastfeeding guidelines or technologies, highlighting the need for tailored training interventions based on experience levels. While nursing is traditionally a female-dominated field, male nurses in pediatric MS settings are increasing. Studies suggest that male nurses may face unique challenges in promoting breastfeeding, often due to perceived cultural barriers or personal discomfort with the subject (Bowdler et al., 2022).

Addressing these gender-based challenges through specific training programs can improve their confidence and ability to support breastfeeding effectively. Higher levels of nursing education are associated with better breastfeeding knowledge and more positive attitudes toward breastfeeding promotion (Khasawneh et al., 2023). Nurses with advanced degrees are more likely to understand the physiological and emotional benefits of breastfeeding, leading to better support for breastfeeding mothers. This highlights the importance of continuous education and professional development for all levels of nursing staff. The number of capacity-building activities nurses participate in also influences their readiness to support breastfeeding. Research shows that nurses who undergo regular breastfeeding-related training are more likely to provide evidence-based breastfeeding support (Gavine et al., 2017). However, in many pediatric MS settings, capacity-building opportunities are either inconsistent or not tailored to the specific challenges faced by nurses in those units. This underscores the need for a structured, frequent, and targeted capacity-building plan to enhance breastfeeding support across all pediatric MS settings (Fok et al., 2022).

3.2 Knowledge, Attitude, and Practices (KAP) Related to Breastfeeding

The KAP framework is widely used to assess the effectiveness of healthcare professionals in promoting health behaviors, such as breastfeeding. For pediatric medical-surgical nurses, their knowledge, attitudes, and practices directly impact how well they support breastfeeding mothers.

Knowledge: Nurses' knowledge of breastfeeding is crucial to providing accurate information and support to mothers. Knowledge gaps among pediatric MS nurses can lead to misinformation, inconsistent advice, and reduced breastfeeding success rates (Čatipović et al., 2022). A systematic review of nursing practices reveals that while most nurses understand the benefits of breastfeeding, they often lack in-depth knowledge of specific techniques, such as managing breastfeeding difficulties or supporting extended breastfeeding (Alakaam et al., 2018).

Attitude: Nurses' attitudes toward breastfeeding are influenced by personal beliefs, cultural norms, and their own experiences with breastfeeding. Positive attitudes toward breastfeeding are essential for promoting its benefits to mothers, but studies show that many nurses still hold reservations about breastfeeding, particularly in medical-surgical settings where breastfeeding may not be prioritized (Smith et al., 2019). Addressing these attitudinal barriers through targeted interventions is critical for improving breastfeeding outcomes.

Practices: Practices refer to how nurses apply their knowledge and attitudes in clinical settings. Inconsistent practices are often noted, with some nurses failing to implement breastfeeding promotion due to time constraints, workload pressures, or lack of institutional support (Sosseh et al., 2023). Research shows that when nurses receive adequate training and institutional backing, their practices align more closely with breastfeeding recommendations, leading to improved breastfeeding rates (Sandhi et al., 2023).

3.3 Comparison of KAP According to Demographic Characteristics

Comparing the KAP of nurses based on demographic factors can provide insights into targeted interventions. For example, studies have found that younger nurses or those with fewer years of experience tend to have lower breastfeeding knowledge but are more open to learning and adopting new practice (Prokop et al., 2021). On the other hand, nurses with more experience may have stronger knowledge but may resist changes in established practices. Gender, as mentioned earlier, also plays a role, with male nurses often feeling less comfortable discussing breastfeeding with patients, potentially due to societal expectations or lack of exposure during training (Wen et al., 2021).

3.4 Relationship Between Knowledge, Attitude, and Practices

The relationship between knowledge, attitudes, and practices (KAP) is complex, and each component influences the others. Studies have consistently shown that nurses with higher levels of breastfeeding knowledge tend to have more positive attitudes and are more likely to engage in best practices when supporting breastfeeding (Ma et al., 2018). Conversely, nurses with knowledge gaps may develop negative attitudes toward breastfeeding or engage in practices that do not support breastfeeding success. Understanding these relationships can help in designing more effective training programs that address all three components simultaneously.

3.5 Capacity Building for Pediatric Medical-Surgical Nurses on Breastfeeding

Capacity building is essential to address the identified gaps in KAP. Pediatric medical-surgical nurses are at the forefront of patient care, and their ability to support breastfeeding has a direct impact on both maternal and infant health outcomes. However, existing breastfeeding training programs in China primarily target general nursing staff and do not specifically address the unique challenges faced by pediatric MS nurses (Liu et al., 2021).

To improve breastfeeding outcomes, a comprehensive capacity-building plan for pediatric MS nurses must be developed. This plan should include regular training on breastfeeding techniques, management of breastfeeding challenges, and strategies to support both exclusive and extended breastfeeding. Additionally, this capacity-building plan should be validated by experts to ensure its relevance and effectiveness in pediatric MS settings.

3.6 Breastfeeding Program in China: Existing Capacity Building Efforts

Breastfeeding promotion has been a key focus of China's national health agenda, particularly with the introduction of the "Outline for the Development of Chinese Children" (2011-2020), which aimed to increase the rate of exclusive breastfeeding. However, most capacity-building efforts have focused on general hospital staff or maternity units, with little attention given to pediatric medical-surgical nurses.

Existing capacity-building initiatives have successfully increased breastfeeding rates in some areas, but they often lack the specialized focus needed for pediatric MS nurses, who face unique challenges in promoting breastfeeding, such as managing infants with complex medical needs (Xin, 2020). A more tailored approach is needed to address these gaps and ensure that pediatric MS nurses are equipped to support breastfeeding effectively.

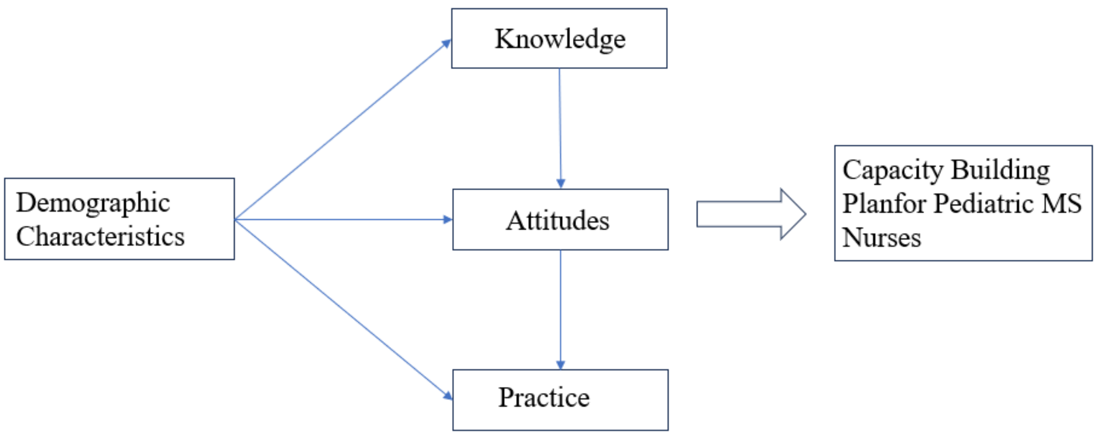
3.7 Future Research Perspectives

Much of the current literature focuses on general nursing staff or neonatal units, with limited research on pediatric MS nurses. While capacity-building programs exist, their long-term impact on breastfeeding rates, particularly in pediatric MS settings, remains under-researched.

3.7.1 Theoretical Framework

This study adopted the Health Belief Model (HBM) as its theoretical foundation to explore the factors and interrelationships within the knowledge, attitudes, and practices (KAP) of pediatric medical-surgical nurses regarding breastfeeding. HBM is a widely applied framework in health behavior research, primarily used to explain how individuals' cognition, attitudes, and beliefs shape their decisions to engage in health-related behaviors (Anuar et al., 2020). In this study, HBM was utilized to analyze how nurses perceived the benefits of breastfeeding, the barriers they may encounter, and their sense of self-efficacy, as well as how these factors influenced their clinical practices.

3.7.2 Conceptual Framework



4. Methods

4.1 Study Design and Locale

This study employed a cross-sectional survey method to comprehensively describe the knowledge, attitudes, and practices (KAP) of respondents regarding neonatal breastfeeding within a specific time frame. Cross-sectional surveys are efficient in terms of time and resources, suitable for large sample sizes, and capable of identifying relationships between variables. Additionally, they provide baseline data for future research. This design is particularly suited to capturing the current state of respondents KAP without the need for long-term follow-up, aligning with the study's goal of providing immediate recommendations for capacity building.

To ensure the homogeneity and representativeness of the study data, this research selected hospitals with well-established pediatric medical-surgical care systems as the study sites. Shenzhen was chosen as the study location due to its hospitals' strong pediatric services and comprehensive breastfeeding education programs integrated into routine care. The study was conducted in four tertiary hospitals in Shenzhen, all of which will remain anonymous to ensure data confidentiality. These hospitals provided a diverse and representative sample of pediatric medical-surgical nurses' knowledge, attitudes, and practices (KAP) regarding neonatal breastfeeding.

The selected hospitals **varied** in size, with bed capacities ranging from 305 to 2,941, and pediatric medical-surgical nursing teams ranging from 25 to 135 nurses. These differences in scale represented a balanced cross-section of healthcare facilities within the same geographic region, ensuring homogeneity while accounting for variations in bed capacity and nursing staff. Such diversity allowed the study to assess differences in resource allocation and staffing practices. The collected data contributed to the development of breastfeeding policies and capacity-building training programs tailored to these healthcare environments.

Table 1. Details of Beds and Pediatric Medical-Surgical Nurses in Hospitals

	Hospital	Beds	Pediatric Medical-Surgical Nurses
1	**Hospital A**	**2941**	**60**

2	**Hospital B**	**600**	**135**
3	**Hospital C**	**830**	**33**
4	**Hospital D**	**305**	**25**

4.2 Study Participants

4.2.1 Sample Size and Sampling

Sample Size Calculation

According to the multi-factor analysis formula n=$4\,\frac{U_{\alpha}^{2}S^{2}}{\delta^{2}}$(Xiao, 2008), the sample size was estimated. The significance level was set at α=0.05, with the corresponding critical value $U_{\alpha/2}$=1.96. Based on previous studies, the S=11.28 (Xin, 2020) and the δ was set as (0.25S,0.5S) (Ni & Zhang, 2011). In this study, "behavior" was selected as the primary outcome indicator for sample size estimation. The rationale for this choice is that behavior is the most direct and critical indicator of nurses' performance in clinical practice, effectively reflecting their actual performance in the workplace. Compared to knowledge and attitude, behavior has a more significant impact on clinical outcomes. Therefore, selecting behavior as the primary outcome helps capture nurses' real-time responses to various nursing tasks, providing stronger support for the development of subsequent intervention measures.

The final calculated sample size ranges from 62 to 246. Based on these calculations and to account for potential variability, the final sample size was set at 150 participants, which falls within the recommended range and provides a sufficient number to ensure robust statistical analysis.

Sampling Method

A proportionate stratified random sampling method was employed to ensure a representative sample of respondents from four anonymized hospitals in Shenzhen, China. First, the total population of 253 Pediatric Medical-Surgical Nurses will be stratified by hospital. The sample size allocated to each hospital was proportionate to the number of respondents in each hospital. Specifically, Hospital A accounts for 23.7% of the total respondent's population, Hospital B for 53.4%, Hospital C for 13.0%, and Hospital D for 9.9%. To achieve the final sample size of 150 participants, the sample from each hospital was adjusted to the nearest whole number: 36 nurses from Hospital A, 80 from Hospital B, 20 from Hospital C, and 14 from Hospital D. Minor rounding adjustments were made to ensure the total sample size of 150 while maintaining representativeness.

Within each hospital, simple random sampling was used to select respondents. Each of population will be assigned a random number, and random numbers will be generated using a random number table or computer-based random number generator to ensure equal selection probability. Recruitment was coordinated with each hospital's nursing administration, and selected respondents were invited to participate via email or phone. Informed consent was obtained from all participants before data collection begins.

To protect the anonymity of participants and smaller hospitals (e.g., Hospital D), all results will be aggregated during data analysis and reporting. Detailed demographic or professional information will not be disclosed in a way that allows identification of individual participants or hospitals. Additionally, specific results from smaller groups will not be reported independently, further safeguarding confidentiality.

4.2.2 Inclusion and Exclusion Criteria

Inclusion criteria:

(1) Actively working registered pediatric medical-surgical nurses in neonatology or pediatric departments were included.

(2) Respondents had successfully completed pre-job training specific to pediatric or neonatal care.

(3) A minimum of 6 months to 1 year of experience in pediatric medical-surgical nursing was required, as this timeframe was regarded as necessary to master basic skills (Kreedi et al., 2021).

(4) Direct experience or formal training in supporting breastfeeding (e.g., hands-on assistance or collaboration with lactation consultants) was required.

(5) Respondents voluntarily agreed to participate in the study after being fully informed about its objectives and procedures.

Exclusion criteria:

(1) Respondents who were on medical leave due to illness.

(2) Respondents who were on leave for educational purposes or were enrolled in full-time educational programs.

(3) Respondents who declined to participate in the study after being fully informed about its objectives, procedures, and potential impact.

Nurses who were on medical leave or pursuing advanced education were excluded from the study, because their unique circumstances did not reflect the typical work conditions of active pediatric medical-surgical nurses, which could have affected the consistency and generalizability of the data. These exclusion criteria were designed to minimize bias and enhance the scientific rigor of the study.

4.3 Research Instruments

Scales were in written form. As participants are Chinese, the language of the surveys used Chinese.

(1) General Information: (APPENDIX 1, section 1)

The questionnaire used a self-developed demographic questionnaire, which included the following items: a) age, b) sex, c) highest educational attainment, d) position category, e) years of nursing experience, f) years of pediatric medical-surgical experience, g) number of capacity building activities within one year.

(2) Questionnaire on breastfeeding knowledge of hospitalized newborns:

(APPENDIX 1, section 2)

The knowledge questionnaire from Yang Piaoyu's study was adapted (Yang & Zhang, 2017). The questionnaire had a content validity index (CVI) of 0.99 and a Cronbach's alpha coefficient of 0.748, indicating good reliability and validity. The questionnaire covered various aspects of breastfeeding, including the benefits of breastfeeding, advocacy, screening, collection, storage, transportation, and breastfeeding operations. It consisted of 20 questions, including 12 single-choice questions and 8 multiple-choice questions. Based on the adapted questionnaire, each correct answer was awarded 5 points, while incorrect answers received 0 points. For multiple-choice questions, full points were only awarded if the selected options matched the reference answer completely; no partial points were given for incomplete or excessive selections. The total score for the questionnaire is 100 points. Higher scores indicated a higher level of breastfeeding knowledge among nurses. Using Bloom's cut-off points, the scores were categorized into three levels:

- Low level: Scores below 60
- Moderate level: Scores between 60 and 80
- High level: Scores above 80.

(3) Questionnaire on breastfeeding attitude of hospitalized newborns:

(APPENDIX 1, section 3)

The attitude questionnaire was adapted from relevant literature, including studies by Wang (2017) and Wang et al. (2018), as well as selected items from the *Evidence-Based Guidelines for Breastfeeding of Hospitalized Newborns*. The questionnaire consisted of 8 questions, using a 5-point Likert scale with the following options: "strongly agree" (5 points), "agree" (4 points), "not sure" (3 points), "disagree" (2 points), and "strongly disagree" (1 point). Based on the adapted questionnaire, the total score for the questionnaire was 40 points. Higher scores reflected a more positive attitude towards breastfeeding among NICU nurses. Using Bloom's cut-off points, the scores were categorized into three levels:

- Low level: Scores below 24
- Moderate level: Scores between 24 and 32
- High level: Scores above 32.

(4) Questionnaire on breastfeeding behavior of hospitalized newborns:

(APPENDIX 1, section 4)

The behavior questionnaire was adapted from the *Evidence-Based Guidelines for Breastfeeding of Hospitalized Newborns*, developed by Zhang Yuxia's team at the Pediatric Hospital affiliated with Fudan University. Based on consultation with clinical experts, five dimensions were selected from the guidelines, covering a total of 28 items. The dimensions included breastfeeding advocacy (3 items, questions 1-3), guidance on the collection and transportation of breast milk (9 items, questions 4-12), screening and receiving breast milk (6 items, questions 13-18), storage, thawing, and heating of breast milk in wards (5 items, questions 19-23), and breastfeeding in wards (5 items, questions 24-28). Each item was scored on a 5-point Likert scale, with the options: "always" (5 points), "often" (4 points), "sometimes" (3 points), "occasionally" (2 points), and "never" (1 point). Questions 22 and 23 were reverse-scored. Based on the adapted questionnaire, the total score for the questionnaire was 140 points. Higher scores indicated better breastfeeding behavior among NICU nurses. Using Bloom's cut-off points, the scores were categorized into three levels:

- Low level: Scores below 84
- Moderate level: Scores between 84 and 112
- High level: Scores above 112.

Pre-survey

Before the formal investigation, a pre-investigation was conducted at four hospitals in Shenzhen, China. Research indicates that for a typical baseline or endline survey, a sample size of approximately 30 to 50 individuals is usually sufficient to identify major issues within the system. Therefore, this study distributed a total of 30 paper-based questionnaires for the pre-experiment (Perneger., 2015). Convenience sampling was used to select participants representing different levels of experience, education, and job roles to ensure a diverse range of perspectives. The purpose of the pre-investigation was to test the rationality of the questionnaire design, evaluate its reliability, and ensure its scientific validity. The questionnaires were distributed in person as paper forms during departmental business meetings at each hospital, with respondents given 20 minutes to complete them. This simulated real-world conditions and assessed the clarity and feasibility of the questionnaire.

Back translation procedures

All questionnaires had received the formal consent of the relevant scale developers, ensuring the legality and ethics of the research. The questionnaires used in this study were adapted from Chinese versions, and the subjects are Chinese individuals; therefore, back-translation of the original questionnaire is not necessary. However, the English questionnaire provided in the appendix underwent back-translation to ensure the accuracy and consistency of the translation.

The back-translation process was carried out as follows: first, the questionnaires were translated from Chinese into English by a professional bilingual translator. Then, a second independent bilingual translator, who was not involved in the original translation, translated the English version back into Chinese. The back-translated Chinese version was compared with the original Chinese version to identify any discrepancies. Any differences were reviewed and resolved to ensure that the English version accurately reflected the meaning of the original Chinese questionnaires.

4.4 Specific Procedures Based on Study Objectives

4.4.1 Preparation Stage

Before the study began, participants were asked to sign a consent form (APPENDIX 2), indicating their willingness to participate in the research. This agreement served as confirmation that the researcher understood the reasons for conducting the study.

4.4.2 Survey Stage

Upon obtaining support from the hospital management department, the survey was scheduled in a way that did not interfere with clinical work. The primary survey times were set in the afternoons after work or on weekends. The location for completing the questionnaires was in a quiet place, such as a departmental study room or an office. Research participants were selected strictly based on the inclusion and exclusion criteria. To minimize bias, all questionnaires were distributed by the researcher in a one-on-one manner, with standardized instructions provided to guide the respondents in completing the questionnaires independently. If participants had any questions about the content of the questionnaire, the researcher provided clarification without influencing their responses, ensuring the reliability of the data collected. The time allotted for completing the questionnaire did not exceed 20 minutes. The questionnaires were distributed, completed, and collected on-site, followed by an immediate review for completeness and logical consistency. If any omissions or obvious logical errors were identified, the participant was asked to correct them on the spot before the questionnaire was officially collected.

4.4.3 Post-Survey Stage

The database for this study was established using Excel 2021, with data entry conducted by the researcher. During the data entry process, the researcher checked for any logical errors, missing values, or outliers in the questionnaires. If the proportion of logical errors, missing values, or outliers exceeded 5%, the questionnaire was considered invalid and excluded from the analysis. After data entry was completed, the researcher randomly selected 20% of the questionnaires for review, comparing the entered data with the original questionnaires to ensure accuracy. If any errors were found during this review, all data were rechecked and corrected. A statistical expert provided guidance during the data analysis phase to ensure the accuracy and scientific rigor of the analysis.

4.5 Ethical Considerations

4.5.1 Informed Consent Process, Duration of Participation, and Withdrawal Criteria

Strict measures were implemented to prevent any perceived coercion during the recruitment process. Recruitment was conducted in collaboration with the hospital's nursing administration to identify eligible participants. However, participants were explicitly informed that their decision to participate, decline, or withdraw from the study at any time would not affect their employment, professional standing, or workplace relationships. All communication emphasized the voluntary nature of the study, ensuring that nurses felt no obligation to participate due to their employer's involvement. Participation in this research study was entirely voluntary, and individuals could choose not to participate or withdraw at any time without any obligations or consequences. Before the study began, the researcher explained all the contents of the informed consent form in detail in a quiet setting, such as a study room, using language that the respondents could understand, avoiding technical jargon, and without inducing or influencing respondents' decisions. The researcher provided the respondents with sufficient time and opportunity to ask questions about the study details and other concerns, allowing them to decide independently whether they wished to participate. The researcher ensured that the respondents fully understood the informed consent form. Both the researcher and the respondents signed and dated the informed consent form on the same date. One copy of the signed and dated form was given to the respondent, while the other copy was retained by the researcher.

4.5.2 Risks and Inconveniences

There was virtually no risk involved in participating in this study, and no identifying information was collected. All data were used solely for this study. On average, each questionnaire took 20 minutes or less to complete. If any of the questions made respondents feel uncomfortable, they could choose not to answer. If you had any questions regarding this study, you could contact Liu Liyue at +86 13631561198 or via email at liu.liyue@auf.edu.ph.

4.5.3 Benefits of the Study

As respondents in this study, there were no associated costs for participation. The study provided an incentive, offering respondents the chance to win a trophy through a lottery. The aim of the study **was** to assess pediatric medical-surgical nurses' knowledge, attitudes, and practices regarding breastfeeding. The findings from this research **could serve** as a foundation for capacity-building programs, helping to enhance nurses' professional competencies in breastfeeding support. This, in turn, **could improve** maternal and infant health, promote better care practices, and reduce the burden on the healthcare system.

4.5.4 Privacy, Confidentiality, and Data Management

Before each respondent registered for this study, the researchers were responsible for providing a comprehensive and detailed introduction to the study's purpose, procedures, and potential risks, and for obtaining a signed written informed consent form. The informed consent form was kept as a clinical research document for future reference. Respondents had the right to withdraw from the study at any time. Upon withdrawal, their personal information and data were destroyed and were not disclosed to any third party. Personal privacy and data confidentiality were protected during the study.

After respondents completed the questionnaire, the research data were collected and managed by the researchers to ensure the accuracy of the experimental data and the privacy of respondents. The original questionnaires were kept by the researchers and stored in a separate locked cabinet. Respondents were coded, and the data were analyzed in a blinded manner, with unresolved issues in the data being reviewed. The stored data did not include the names of respondents, meaning that the published data analysis could not be traced back to them. All other relevant information, including answers to questions in the survey, was used solely for research purposes. The actual survey forms and summary tables were kept for three years after the study concluded, after which they were destroyed using a shredder, and a final check was conducted to ensure that all data and questionnaires in the drawer had been completely destroyed. The results of all questionnaires were not used directly or indirectly for any other research.

4.5.5 Conflict of Interest

This research was not funded, and the researcher did not see any conflict of interest in this work.

5. Statistical Analysis of Data

Data were analyzed using SPSS 29.0 software. The numerical data were described using mean and standard deviation for normally distributed data, and median and interquartile range for non-normally distributed data. Categorical data were described using frequency and percentage. The normality of the knowledge, attitude, and behavior scores was assessed using the Shapiro-Wilk test. Scatter plots were also used to determine any linear relationships between variables.

For the comparison of KAP scores across different demographic groups, independent samples t-tests were employed for dichotomous variables (e.g., gender), and one-way ANOVA was used for variables with more than two categories (e.g., educational attainment) when the data were normally distributed. If the data did not meet the normality assumption, the Mann-Whitney U test was used for comparing two groups, while the Kruskal-Wallis test was applied for comparisons involving three or more groups.

To examine relationships between knowledge, attitudes, and practices, Pearson's correlation was used for normally distributed data, while Spearman's rank correlation was applied for non-normally distributed data. Correlation strengths were interpreted as very weak (0.00–0.19), weak (0.20–0.39), moderate (0.40–0.59), strong (0.60–0.79), and very strong (0.80–1.00).

All p-values were two-sided, and statistical significance was set at $p < 0.05$.

6. Results

The demographic characteristics of the participants indicate that the majority of respondents were female (95.13%), with a significantly smaller proportion being male (4.87%). This gender imbalance aligns with global nursing workforce trends, where females dominate the profession. For instance, the World Health Organization (WHO) reports that over 67.2% of nurses worldwide are female, reflecting historical and sociocultural factors influencing career choices in healthcare (WHO, 2020). In terms of educational attainment, most participants had a bachelor's degree (80.97%), while a smaller percentage held qualifications below a bachelor's degree (18.58%) or a master's degree and above (0.44%). This pattern may be linked to professional thresholds and career advancement mechanisms in nursing. For instance, the high proportion of bachelor's degrees (80.97%) could reflect hospitals' increasing educational requirements for clinical nurses, while the low rate of master's degrees (0.44%) may align with frontline roles prioritizing practical experience over academic credentials (Dall'Ora et al., 2022). Regarding capacity-building activities, the majority (63.27%) attended one to two activities in the past year, while a smaller proportion attended three to five activities (13.72%) or more than six activities (10.18%). A notable 12.83% of participants did not attend any capacity-building activities. This disparity may reflect uneven access to training resources or varying institutional support. Prior studies highlight that nurses in resource-constrained settings often face barriers to continuing education, such as heavy workloads or lack of funding (Mlambo et al., 2021). The numerical characteristics reveal that the average age of participants was 30.76 years (SD = 5.79), with an average of 9.15 years (SD = 5.90) of nursing experience and 8.10 years (SD = 5.59) of medical-surgical experience.

Table 2. Summary of the demographic characteristics of the participants

Characteristics	Categories	Frequency	Percentage
Gender	Female	215	95.13 %
	Male	11	4.87%
Educational Attainment	Below Bachelor's Degree	42	18.58 %
	Bachelor's Degree	183	80.97 %

	Master's Degree and above	1	0.44 %
Number of Capacity Building Activities Within One Year	1-2 times	143	63.27 %
	3-5 times	31	13.72 %
	More than 6 times	23	10.18 %
	None	29	12.83 %
Numerical Characteristics	**Mean**	**Median**	**Standard deviation**
Age	30.76	30.00	5.79
Nursing experience	9.15	9.00	5.90
Medical-surgical experience	8.10	8.00	5.59

Table 2 reveals significant disparities in breastfeeding knowledge among participants. While foundational concepts like colostrum functions (71.24%) and WHO-recommended exclusive breastfeeding duration (51.33%) were moderately understood, critical gaps emerged in clinical and policy-related areas: only 1.77% recognized the WHO/UNICEF breastfeeding initiative, 24.78% identified preterm infant fortification criteria, and 11.95% understood contamination reduction practices. Conversely, practical scenarios such as discarding unused heated milk (97.35%) and labeling stored milk (92.92%) showed high accuracy. Combined with Table 3's findings—88.05% scoring low (mean = 46.02/100)—these results underscore an urgent need for training focused on evidence-based guidelines (e.g., WHO, 2023) and clinical protocols to address underrecognized topics.

Table 3. Summary of the knowledge of participants related to breastfeeding

Item	Correct Answers	
	Frequency	**Percentage**
Identify a function that is not considered important for breastfeeding.	115	50.88 %
Up to what age does the WHO recommend exclusive breastfeeding?	116	51.33 %
According to China's guidelines, preterm infants with a gestational age of how many weeks or less and a birth weight of how many grams or less should primarily receive fortified breast milk?	56	24.78 %
Mothers with which diseases should avoid breastfeeding?	168	74.34 %
What is not a basic principle for using medication while breastfeeding?	159	70.35 %
How is colostrum defined in terms of the time frame after birth?	84	37.17 %
To increase milk production, how often should a mother pump, and for how many minutes each time?	18	7.96 %
Identify an incorrect statement about breast milk storage.	75	33.19 %
What is the best method for thawing frozen breast milk?	104	46.02 %
At what temperature (°C) should thawed breast milk be warmed in water?	72	31.86 %
What should be done with thawed breast milk that has been heated but not consumed?	220	97.35 %
Identify an incorrect statement about breastfeeding practices.	205	90.71 %
Which initiative was formulated by WHO and UNICEF to protect, support, and promote breastfeeding?	4	1.77 %

What are some measures to promote breastfeeding in hospitalized infants?	190	84.07 %
What are the functions of colostrum?	161	71.24 %
Identify an incorrect statement about medication use during breastfeeding.	6	2.65 %
When labeling breast milk sent to a hospital, what additional information should be included besides the bed number and name?	210	92.92 %
What are effective methods to stimulate the let-down reflex?	45	19.91 %
What is a correct practice to reduce contamination during breast milk collection?	27	11.95 %
Which statement about breast milk transport is correct?	45	19.91 %

The overall knowledge assessment (Table 3) highlights a significant deficit in breastfeeding expertise among participants, with a mean score of 46.02/100 (SD = 10.30) indicating a widespread lack of comprehensive understanding. Notably, 88.05% of participants scored in the low-knowledge range (5–59), while only 11.95% attained moderate proficiency (60–80), and none achieved high knowledge levels (81–100). This pronounced knowledge gap is particularly concerning given the critical role of healthcare providers in promoting breastfeeding practices (Smith et al., 2018). The high standard deviation (SD = 10.30) suggests considerable variability in knowledge levels, potentially reflecting disparities in prior training or exposure to breastfeeding guidelines. These findings align with specific deficiencies identified in Table 3, such as the minimal awareness of WHO/UNICEF initiatives (1.77%), underscoring the urgent need for targeted educational interventions. Previous studies have demonstrated that structured training programs focusing on evidence-based guidelines and policy awareness can significantly enhance breastfeeding knowledge and clinical competency (Bowdler et al., 2022). For instance, Khasawneh et al. (2023) found that nurses with higher knowledge scores were more effective in supporting breastfeeding mothers, leading to improved infant health outcomes.

Table 4. Descriptive statistics of the total knowledge of participants related to breastfeeding

Statistic / Category	**Value / Score Range**	**Frequency**	**Percentage**
Mean	46.02	-	-
Median	45.00	-	-
Standard deviation	10.30	-	-
Minimum	20	-	-
Maximum	80	-	-
Low level of knowledge	5 – 59	199	88.05 %
Moderate level of knowledge	60 – 80	27	11.95 %
High level of knowledge	81 – 100	0	0.00 %

The attitude assessment (Table 5) revealed strong consensus among participants regarding the value of breastfeeding for pediatric medical-surgical infants. Across all items, mean scores ranged from 4.5 to 4.7 (out of 5), indicating uniformly positive perceptions. A striking 97.35% of respondents strongly agreed (SA) or agreed (A) that nurses play a crucial role in breastfeeding support (M=4.7, SD=0.58). Similarly, 95.58% endorsed the superiority of direct breastfeeding over expressed milk (M=4.62, SD=0.64*M*=4.62, *SD*=0.64), consistent with WHO guidelines advocating for maternal-infant bonding to optimize immunological benefits (Victora et al., 2016).

Paradoxically, despite high confidence in their ability to support breastfeeding mothers (M=4.66, SD=0.61), participants simultaneously acknowledged significant gaps in institutional training (M=4.5, SD=0.76), with 91.15% advocating for enhanced educational initiatives. This discrepancy mirrors findings by Najafi and Nasiri (2023), who identified a pervasive "competency-confidence gap" among nurses lacking updated clinical knowledge.

Table 5. Summary of the attitude of participants related to breastfeeding

Items	Mean	SD	Frequency (Percentage)				
			SDA	D	N	A	SA
I believe that breastfeeding directly by mothers is better than expressing milk for Pediatric Medical-Surgical babies	4.62	0.64	2 (0.88%)	0 (0.00%)	8 (3.54%)	61 (26.99%)	155 (68.58%)
I believe that breastfeeding is beneficial for Pediatric Medical-Surgical newborns	4.71	0.58	2 (0.88%)	0 (0.00%)	2 (0.88%)	54 (23.89%)	168 (74.34%)
I think mothers need more help from nurses to successfully breastfeed Pediatric Medical-Surgical babies	4.68	0.59	2 (0.88%)	0 (0.00%)	3 (1.33%)	58 (25.66%)	163 (72.12%)
I believe that the role of nurses is crucial in the breastfeeding of Pediatric Medical-Surgical babies	4.7	0.58	2 (0.88%)	0 (0.00%)	2 (0.88%)	56 (24.78%)	166 (73.45%)
I believe breastfeeding is better than formula feeding for Pediatric Medical-Surgical babies	4.65	0.62	2 (0.88%)	0 (0.00%)	6 (2.65%)	58 (25.66%)	160 (70.80%)
I am confident in my ability to support mothers in breastfeeding their Pediatric Medical-Surgical babies	4.66	0.61	2 (0.88%)	0 (0.00%)	5 (2.21%)	59 (26.11%)	160 (70.80%)
Our unit provides adequate support and resources for breastfeeding Pediatric Medical-Surgical babies	4.6	0.64	2 (0.88%)	0 (0.00%)	7 (3.10%)	68 (30.09%)	149 (65.93%)
I think more training on breastfeeding Pediatric Medical-Surgical babies is needed for nurses	4.5	0.76	2 (0.88%)	3 (1.33%)	15 (6.64%)	65 (28.76%)	141 (62.39%)

Note: SD= Standard deviation; SDA=Strongly Disagree; D= Disagree; N=Not sure; A=Agree; SA=strongly agree.

Table 6 summarizes the attitudes of participants toward breastfeeding, revealing a predominantly positive stance. The mean attitude score was 37.13 (SD = 4.62), with a median of 40.00 (maximum possible score = 40), indicating a strong skew toward high attitudinal endorsement. Notably, 76.11% of participants scored in the high-attitude range (33–40), while only 0.88% exhibited low attitudes (8–23). The narrow standard deviation (4.62) suggests minimal variability in responses, reflecting a consensus on the importance of breastfeeding. However, the discrepancy between the mean (37.13) and median (40.00) points to a potential left-skewed distribution, possibly due to a small number of lower scores (e.g., minimum = 8).

Table 6. Descriptive statistics of the total attitude of participants related to breastfeeding

Statistic / Category	Value / Score Range	Frequency	Percentage
Mean	37.13	-	-
Median	40.00	-	-
Standard deviation	4.62	-	-
Minimum	8	-	-
Maximum	40	-	-
Low level of attitude	8 – 23	2	0.88 %
Moderate level of attitude	24 – 32	52	23.01 %
High level of attitude	33 – 40	172	76.11 %

Table 7 highlighted the extensive breastfeeding-related practices among healthcare providers, particularly in pediatric medical-surgical settings. The results indicated a strong emphasis on breastfeeding advocacy, as evidenced by the high mean scores for providing knowledge about breastfeeding (M = 4.14, SD = 1.00) and emphasizing its importance to families (M = 4.35, SD = 0.86). Additionally, the guidance provided for the collection and transportation of breast milk was well-practiced, with notable adherence to hygiene protocols, such as washing hands before milk collection (M = 4.36, SD = 0.96) and maintaining proper storage conditions (M = 4.50, SD = 0.78). Screening and receiving breast milk also showed high compliance, particularly in verifying labels (M = 4.59, SD = 0.71) and ensuring the safety of stored milk (M = 4.58, SD = 0.76). However, certain aspects, such as heating breast milk using a warm water bath (M = 2.46, SD = 1.75) and freezing leftover milk after feeding (M = 2.42, SD = 1.75), received relatively lower scores, indicating areas for improvement. Moreover, breastfeeding-related practices in wards were well-maintained, particularly in verifying the identity of mothers and babies (M = 4.37, SD = 0.89) and monitoring feeding progress across wards (M = 4.65, SD = 0.69).

Table 7. Summary of the practices of participants related to breastfeeding

Items	Mean	SD	Frequency (Percentage)				
			N	OC	S	OF	A
Breastfeeding advocacy							
I provide handouts or brochures to families of Pediatric Medical-Surgical patients to monitor breast milk collection	3.94	1.16	11 (4.87%)	20 (8.85%)	33 (14.60%)	69 (30.53%)	93 (41.15%)
I emphasize the importance of breastfeeding to the families of Pediatric Medical-Surgical patients	4.35	0.86	2 (0.88%)	7 (3.10%)	25 (11.06%)	69 (30.53%)	123 (54.42%)
I actively provide knowledge about breastfeeding (such as teaching and demonstrating techniques, providing supporting equipment, etc.),	4.14	1	5 (2.21%)	11 (4.87%)	36 (15.93%)	69 (30.53%)	105 (46.46%)

creating an encouraging environment to promote the initiation of breastfeeding							
Guidance on the collection and transportation of breast milk							
I guide families of Pediatric Medical-Surgical patients to collect breast milk within six hours after birth	3.81	1.31	23 (10.18%)	14 (6.19%)	40 (17.70%)	56 (24.78%)	93 (41.15%)
I guide families to express milk 2-3 times per day	4.05	1.1	9 (3.98%)	13 (5.75%)	38 (16.81%)	63 (27.88%)	103 (45.58%)
I guide families to wash their hands thoroughly before each breast milk collection	4.36	0.96	6 (2.65%)	6 (2.65%)	23 (10.18%)	56 (24.78%)	135 (59.73%)
I guide families to clean the pumping room (without using disinfectants to avoid skin irritation)	4.27	1	7 (3.10%)	7 (3.10%)	26 (11.50%)	63 (27.88%)	123 (54.42%)
I guide families on cleaning and disinfecting breast milk collection equipment	4.28	0.96	5 (2.21%)	8 (3.54%)	26 (11.50%)	66 (29.20%)	121 (53.54%)
I guide families to store breast milk separately each time	4.41	0.88	3 (1.33%)	6 (2.65%)	23 (10.18%)	58 (25.66%)	136 (60.18%)
I guide families to practice good hygiene when storing breast milk	4.5	0.82	3 (1.33%)	5 (2.21%)	15 (6.64%)	57 (25.22%)	146 (64.60%)
I guide families to discard the first drops of naturally collected milk	4.08	1.19	15 (6.64%)	12 (5.31%)	24 (10.62%)	63 (27.88%)	112 (49.56%)
I guide families to transport breast milk using coolers or dry ice, maintaining the cold chain during transportation	4.5	0.78	1 (0.44%)	7 (3.10%)	13 (5.75%)	61 (26.99%)	144 (63.72%)
Screening and receiving breast milk							
I consult with physicians about medications taken by mothers to ensure they do not affect breastfeeding	4.33	0.92	6 (2.65%)	5 (2.21%)	20 (8.85%)	72 (31.86%)	123 (54.42%)
I consult with physicians about mothers' illnesses to ensure they do not affect breastfeeding	4.34	0.93	7 (3.10%)	3 (1.33%)	20 (8.85%)	72 (31.86%)	124 (54.87%)
When accepting breast milk, I check whether the labels are intact and clear (including date and time of collection)	4.59	0.71	1 (0.44%)	3 (1.33%)	15 (6.64%)	50 (22.12%)	157 (69.47%)
When accepting breast milk, I check the volume and quality	4.58	0.71	1 (0.44%)	3 (1.33%)	14 (6.19%)	54 (23.89%)	154 (68.14%)
When accepting breast milk, I sign according to the	4.61	0.71	2 (0.88%)	1 (0.44%)	14 (6.19%)	50 (22.12%)	159 (70.35%)

regulations							
When accepting breast milk, I store it immediately for later use	4.58	0.76	3 (1.33%)	2 (0.88%)	13 (5.75%)	51 (22.57%)	157 (69.47%)
Storage, thawing, and heating of breast milk in wards							
I check and clean the breast milk freezer in the ward every day	4.54	0.75	2 (0.88%)	3 (1.33%)	15 (6.64%)	57 (25.22%)	149 (65.93%)
I separate and label each mother's breast milk in the freezer	4.6	0.73	2 (0.88%)	2 (0.88%)	14 (6.19%)	49 (21.68%)	159 (70.35%)
I use breast milk according to the order of collection, prioritizing colostrum and fresh breast milk	4.57	0.73	1 (0.44%)	3 (1.33%)	17 (7.52%)	50 (22.12%)	155 (68.58%)
I heat breast milk using a warm water bath	2.46	1.75	126 (55.75%)	6 (2.65%)	13 (5.75%)	27 (11.95%)	54 (23.89%)
I freeze leftover breast milk after feeding	2.42	1.75	131 (57.96%)	1 (0.44%)	14 (6.19%)	27 (11.95%)	53 (23.45%)
Breastfeeding in wards							
When checking in new patients, I verify the identity and labels of the mother and baby	4.37	0.89	2 (0.88%)	9 (3.98%)	24 (10.62%)	60 (26.55%)	131 (57.96%)
When checking in new patients, I check the milk for air bubbles	4.4	0.98	7 (3.10%)	7 (3.10%)	18 (7.96%)	51 (22.57%)	143 (63.27%)
When checking in new patients, I check the color of the breast milk	4.57	0.73	2 (0.88%)	2 (0.88%)	14 (6.19%)	56 (24.78%)	152 (67.26%)
During breastfeeding in the ward, I avoid exposing the milk to light	4.21	1.2	19 (8.41%)	3 (1.33%)	21 (9.29%)	51 (22.57%)	132 (58.41%)
I pay attention to the feeding progress of babies in other wards as well	4.65	0.69	2 (0.88%)	1 (0.44%)	12 (5.31%)	45 (19.91%)	166 (73.45%)

Table 8 indicates a strong adherence to recommended breastfeeding practices among healthcare providers, with a mean practice score of 118.50 (SD = 18.55) out of a possible 140. The majority of participants (61.95%) exhibited high levels of practice proficiency (113–140), while only 4.42% scored in the low range (28–83). This high performance aligns with institutional protocols and standardized workflows, as seen in Table 6's rigorous compliance with hygiene practices (e.g., 64.60% "always" instructed proper storage). However, the significant standard deviation (18.55) and minimum score of 48 suggest variability, potentially linked to gaps in complex clinical scenarios requiring advanced knowledge (e.g., preterm infant fortification in Table 2, 24.78% accuracy).

These findings resonate with studies showing that structured clinical guidelines can drive high compliance in routine tasks, yet knowledge deficits may undermine adaptability in non-routine situations (Najafi & Nasiri, 2023). For instance, while participants excelled in daily freezer maintenance (Table 6, M=4.54), their lower accuracy in applying colostrum prioritization (M=4.57) reflects a need for deeper integration of evidence-based principles.

Table 8. Descriptive statistics of the total practices of participants related to breastfeeding

Statistic / Category	Value / Score Range	Frequency	Percentage
Mean	118.50	-	-
Median	122.00	-	-
Standard deviation	18,55	-	-
Minimum	48	-	-
Maximum	140	-	-
Low level of practices	28 – 83	10	4.42 %
Moderate level of practices	84 – 112	76	33.63 %
High level of practices	113 – 140	140	61.95 %

Table 9 provided insights into the factors influencing breastfeeding-related practices. Notably, educational attainment showed a significant association with knowledge (p = 0.0476), indicating that participants with a bachelor's degree or higher had greater breastfeeding-related knowledge than those with lower educational attainment, suggesting formal education enhanced theoretical understanding of clinical guidelines, as supported by Pereira et al. (2022). Moreover, age (p = 0.0185) and nursing experience (p = 0.0267) were positively correlated with knowledge, suggesting that more experienced and older healthcare providers possessed greater breastfeeding-related knowledge. In terms of practices, the frequency of engagement in breastfeeding training or education was significantly related (p = 0.0024), with those attending training more frequently exhibiting better practices. Additionally, age (p = 0.0030) and nursing experience (p = 0.0013) were significantly associated with higher levels of breastfeeding practices, implying that experience and professional development played key roles in fostering effective breastfeeding-related practices.

Table 9. Comparison and relationship of knowledge, attitude, and practices (KAP) related to breastfeeding and demographic characteristics

Demographic Characteristics	Knowledge		Attitude		Practices	
	Median	p-value	Median	p-value	Median	p-value
Female	45.00	0.3383	37.20	0.1064	118.90	0.5313
Male	45.00		35.73		110.82	
Bachelor's Degree and Up	46.60	*0.0476	37.41	0.1637	119.35	0.2683
Below Bachelor's Degree	43.45		35.90		114.79	
1-2 times	45.00	0.4201	39.00	0.0758	117	*0.0024
3-5 times	45.00		40.00		132	
More than 6 times	45.00		40.00		132	
None	45.00		40.00		114	
Age	r = 0.16	*0.0185	r = 0.12	0.0717	r = 0.20	*0.0030
Nursing experience	r = 0.15	*0.0267	r = 0.10	0.1237	r = 0.21	*0.0013
Medical-surgical experience	r = 0.09	0.1922	r = 0.10	0.1463	r = 0.17	*0.0114

Note: The Mann-Whitney U test was used for categorical variables with two groups, the Kruskal-Wallis test was applied to categorical variables with more than two groups, and Spearman's correlation was used for numerical variables. * - significant at p-value less than 0.05.

Table 10 examines the correlations between breastfeeding-related knowledge, attitudes, and practices (KAP) using Spearman's rank correlation. The results reveal a nuanced interplay among these dimensions. First, **knowledge** showed no significant association with **attitude** (ρ=−0.04, p=0.6004), suggesting that higher theoretical understanding does not inherently translate to more positive attitudes. This aligns with prior research (Hu et al., 2021). For instance, while participants demonstrated substantial knowledge gaps in critical areas like preterm infant fortification (24.78% accuracy, Table 2), their self-reported confidence in breastfeeding support remained high (Table 4, M=4.66), implying that institutional advocacy or workplace culture may override individual knowledge limitations.

Similarly, knowledge exhibited no meaningful correlation with practices (ϱ=−0.07, p=0.2898), highlighting a systemic disconnect between theoretical guidelines and their clinical application. For example, despite strong adherence to routine protocols such as milk labeling (M=4.59, Table 6), participants struggled with complex tasks like contamination reduction (11.95% accuracy, Table 2), underscoring the need for training that bridges abstract knowledge to actionable workflows.

In contrast, **attitude** and **practices** displayed a robust positive correlation (ϱ=0.54, p<0.0001), emphasizing that proactive attitudes are a critical driver of protocol adherence. This finding resonates with the Theory of Planned Behavior, where attitudinal commitment directly predicts behavioral outcomes (Verplanken & Orbell, 2021). The high practice scores observed in Table 7 (mean = 118.50/140) may thus reflect the pervasive positive attitudes reported in Table 4 (e.g., 73.45% strongly agreeing on nurses' crucial role).

Table 10. Correlation matrix of knowledge, attitude, and practices (KAP) related to breastfeeding of participants

Correlation Matrix		Knowledge	Attitude	Practices
Knowledge	Spearman's rho	—		
	df	—		
	p-value	—		
Attitude	Spearman's rho	-0.04	—	
	df	224	—	
	p-value	0.6004	—	
Practices	Spearman's rho	-0.07	0.54	—
	df	224	224	—
	p-value	0.2898	*< .0001	—

Note: Spearman's correlation was used. * - significant at p-value less than 0.05.

7. Discussion

7.1 Knowledge and Practice Disconnection

The results of this study show a significant imbalance in the pediatric medical-surgical nurses' knowledge of breastfeeding. While basic knowledge is relatively well understood, there are significant gaps in key areas. For example, while most nurses understand the basic principles of breastfeeding, only half of them can correctly identify the World Health Organization (WHO) recommendation for exclusive breastfeeding for the first six months. Additionally, the accuracy rate for questions related to the latest guidelines and professional knowledge, such as fortification criteria for preterm infants and milk storage methods, is relatively low.

This knowledge gap directly affects the nurses' practical behavior. The study found that although most nurses recognize the importance of breastfeeding, the frequency of actively supporting breastfeeding in clinical practice is still low. Few nurses regularly provide guidance on proper breastfeeding techniques or develop lactation support plans for mothers. This finding aligns with Van De Ven and Johnson (2006), who observed that nurses, despite having certain theoretical knowledge, often encounter difficulties in translating this knowledge into practice.

There are several reasons for the disconnect between knowledge and practice. From the individual level, nurses' confidence and competence are closely related to their knowledge base (Pueyo-Garrigues et al., 2021). If nurses are unfamiliar with scientific feeding plans or handling techniques, even with a positive attitude, they may refrain from taking action due to lack of confidence. Moreover, long-term work habits and departmental divisions may lead nurses to focus more on disease treatment and neglect breastfeeding support, creating a "treatment-heavy, feeding-light" trend. Similar findings have been reported in studies from other countries (Shakhshir & Alkaiyat, 2023). Studies have shown that nurses, although possessing basic breastfeeding knowledge, fail to actively intervene in practice due to high work pressure and lack of systematic training.

At the institutional level, the failure to incorporate breastfeeding into routine nursing processes and performance evaluation systems is a major factor contributing to the disconnect between knowledge and practice. If hospitals fail to provide necessary support and incentive mechanisms, nurses may have the knowledge but lack the motivation and conditions to apply it in practice (Zeng et al., 2022).

7.2 Tension Between Attitudes and Institutional Support

This study also found that while nurses generally hold positive attitudes towards breastfeeding, recognizing its importance for infant health, these attitudes have not consistently translated into practice. This reflects the significant tension between personal attitudes and institutional support. On one hand, positive attitudes are a driving force for breastfeeding support: nurses recognize the importance of breastfeeding for infant health and are willing to provide emotional support to mothers. On the other hand, a lack of systematic institutional support becomes a major obstacle to translating these attitudes into action.

For example, some nurses in this study reported that their departments lacked an environment that encouraged breastfeeding, and the hospital did not provide sufficient resources, such as lactation rooms or personnel to assist mothers with breastfeeding. In such cases, even if nurses have a positive attitude, they may not be able to put it into action due to the lack of institutional support. This finding is consistent with Meek and Noble (2022), who noted that despite nurses globally recognizing the benefits of breastfeeding, the lack of systemic support led to a failure in translating knowledge into practice.

Moreover, the lack of institutional support may also negatively impact nurses' attitudes. When hospital management does not prioritize breastfeeding, nurses may lower their perception of its importance. Some nurses may not see breastfeeding support as part of their core responsibilities, especially in pediatric wards, where the focus is more on medical care, and feeding support is seen as secondary. To address this issue, hospitals need to strengthen institutional support to ensure that nurses' attitudes and their practical work align.

8. Conclusion

Low Level of Breastfeeding Knowledge: This study found that pediatric nurses have a relatively low level of breastfeeding knowledge, particularly in areas such as preterm infant fortification and milk storage. Nurses generally have a basic understanding of breastfeeding, but their knowledge of the latest guidelines and detailed practices is limited.

Positive Attitudes but Insufficient Practice: While most nurses have a positive attitude toward breastfeeding and recognize its importance for infant health, this attitude has not been fully translated into consistent practice. The frequency of support for breastfeeding in daily clinical work is still inadequate.

Disconnect Between Knowledge and Practice: There is a disconnect between the knowledge nurses possess and the actions they take in practice. Some nurses, despite having high levels of theoretical knowledge, fail to actively support breastfeeding due to lack of hands-on experience and institutional support.

Tension Between Attitudes and Institutional Support: Although nurses generally support breastfeeding, the lack of institutional support has hindered their ability to implement breastfeeding practices effectively. This indicates that personal attitudes alone are insufficient without the support of a strong institutional framework.

Multiple Factors Influence KAP: Both personal factors (such as education level and training experience) and institutional factors (such as hospital policies and training opportunities) jointly influence nurses' knowledge, attitudes, and practices related to breastfeeding.

9. Recommendation

9.1 Nurse Continuing Education and Training

Strengthening the continuing education system for pediatric nurses on breastfeeding is crucial to filling knowledge gaps and improving practical abilities. This study identified significant gaps in new breastfeeding knowledge and specialized skills, which need to be addressed through systematic training. Hospitals and relevant departments should develop long-term training programs, incorporating breastfeeding knowledge and skills as key components of continuing education for nurses. Training content should include the latest international and national guidelines, feeding strategies for special populations (such as preterm infant fortification), standards for milk storage and handling, and solutions to common breastfeeding issues, with a focus on the areas highlighted in this study. Training methods should include lectures, workshops, and simulated practice to increase the practical applicability and participation rate.

To ensure the effectiveness of the training, it is recommended to introduce assessment and feedback mechanisms. Nurses should undergo knowledge tests and practical evaluations after training, with results linked to performance reviews to encourage practical application of the learned content. Hospital management should support and reward nurses who participate in breastfeeding-related certifications (e.g., International Lactation Consultant Certification), fostering specialized talent. Additionally, WHO/UNICEF-developed breastfeeding support courses could be adopted, with experienced lactation consultants or nutritionists providing evidence-based guidance. This ongoing education will gradually improve pediatric nurses' knowledge and skills, laying a solid foundation for improving clinical practice.

9.2 Hospital System Construction

Hospitals should create an environment that supports breastfeeding and integrate breastfeeding support into routine pediatric nursing practices. This study reveals that the lack of institutional support is one of the main reasons for insufficient breastfeeding practice, so hospital management needs to take proactive measures to improve related systems. First, hospitals can establish clear breastfeeding support protocols and processes, such as requiring pediatric nurses to assess breastfeeding conditions upon patient admission, provide appropriate guidance, and regularly inquire about and assist mothers' breastfeeding needs during hospitalization. Incorporating these measures into routine nursing practices will help make breastfeeding support an automatic part of every nurse's duties.

Hospitals should provide necessary hardware and resources, such as setting up breastfeeding rooms or private spaces, providing breast pumps, refrigeration equipment, etc., to facilitate mothers' breastfeeding and milk storage. For hospitalized infants who cannot breastfeed directly, a milk storage and feeding system should be established to ensure that breast milk can be safely collected, stored, and fed to the infant.

Furthermore, hospital management should encourage departments to create an atmosphere that supports breastfeeding, integrating it into departmental quality management and performance assessments. Multidisciplinary discussions, where pediatric nurses collaborate with pediatricians, nutritionists, and others, should be held regularly to ensure breastfeeding takes precedence in patient care. Departments and individuals who perform well in supporting breastfeeding can be rewarded as role models. Hospitals can also refer to the Baby-Friendly Hospital Initiative and implement measures that support breastfeeding, such as allowing mother-infant rooming-in or extending mother accompaniment time to minimize unnecessary formula feeding. These institutional reforms will help alleviate nurses' concerns about supporting breastfeeding, enabling them to actively implement practices under the drive of their positive attitudes and reducing the current knowledge-practice gap.

9.3 Policy Guidance

Health administrative departments and industry associations should play a macro role in guiding the improvement of healthcare providers' breastfeeding support capabilities from a policy perspective. First, it is recommended to develop and improve breastfeeding guidelines or technical specifications for healthcare personnel, incorporating pediatric nurses, and clearly defining their responsibilities in breastfeeding support. These guidelines should be issued to healthcare institutions as industry standards and regularly updated to include the latest evidence-based medical information.

Health authorities should strengthen the supervision and assessment of breastfeeding support practices in healthcare institutions. Indicators such as the breastfeeding rates of hospitalized infants, whether hospitals have established related policies, and nurse training participation rates should be included in hospital evaluation and grading systems, urging hospital administrators to pay attention and take action. This top-down pressure will turn into motivation at the grassroots level, pushing hospitals to implement nurse training and system building.

9.4 Future Research

Although this study has explored the current state of pediatric nurses' breastfeeding KAP, several areas remain for further research. First, similar surveys should be conducted in a wider range of regions and populations to test the generalizability of the findings. Future studies can include a larger sample of pediatric nurses across different provinces and cities, comparing differences between regions and hospital levels and analyzing how influencing factors operate in different contexts. This will help design targeted regional policies and training programs.

Qualitative research should be conducted to complement the quantitative findings. In-depth interviews with pediatric nurses can reveal their subjective reasons for supporting or failing to support breastfeeding in their work, such as work pressure, confidence in knowledge, and interpersonal factors, enriching the understanding of factors influencing KAP. Similarly, interviews with mothers or caregivers of hospitalized children can provide feedback on their needs and evaluations of nurse support, offering insights for improving nurse practices.

Intervention studies are encouraged, applying the strategies proposed in this research and evaluating their effectiveness. For example, breastfeeding training programs for pediatric nurses or department support plans could be implemented in a group of hospitals, with a control group of hospitals that have not implemented these interventions, and comparisons can be made after some time to evaluate the changes in nurse KAP levels and patient breastfeeding outcomes. These studies will provide empirical evidence to confirm the effectiveness of training and institutional interventions, further persuading policymakers to invest more resources. Finally, future research could expand into interdisciplinary collaboration, involving clinical medicine, nursing management, public health, and other fields to comprehensively assess and improve nurses' breastfeeding support capacity.

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Appendix 1

Section 1: General Information Questionnaire

No.	Item	Options
A01	Age (years)	( ) years
A02	Sex	(1) Male (2) Female
A03	Highest Educational Attainment	(1) Below Bachelor's Degree (2) Bachelor's Degree (3) Master's Degree and above
A04	Position Category	(1) Nurse (2) Nurse Practitioner (3) Head Nurse or above
A05	Years Of Nursing Experience	( ) years
A06	Years Of Pediatric Medical-Surgical Experience	( ) years
A07	Number Of Capacity Building Activities Within One Year	( )

Section 2: Questionnaire on Pediatric Medical-Surgical Nurses' Knowledge of Breastfeeding for Hospitalized Newborns

The following items are a survey of Pediatric Medical-Surgical nurses' knowledge on breastfeeding for hospitalized newborns. Questions 1-12 are single-choice, and questions 13-20 are multiple-choice. Please fill in the correct options you think are appropriate in the brackets after the questions.

B01. Which of the following is not an important function of breastfeeding: ( )

A. Promotes brain development and long-term developmental outcomes

B. Reduces feeding intolerance

C. Reduces uterine contraction in mothers

D. Prevents infections

B02. WHO recommends exclusive breastfeeding up to the age of: ( )

A. 4 months

B. 6 months

C. 9 months

D. 12 months

B03. According to China's "Feeding Recommendations for Preterm/Low Birth Weight Infants," preterm infants with a gestational age of ≤ _ weeks and a birth weight of ≤ _ grams should primarily receive fortified breast milk: ( )

A. 35; 1500

B. 36; 2000

C. 34; 2000

D. 35; 1000

B04. Mothers with which of the following diseases should not breastfeed: ( )

A. Hypothyroidism treated with thyroid hormone postpartum

B. Hepatitis B with minor symptoms

C. Syphilis

D. Undergoing cytotoxic chemotherapy

B05. Which of the following is not a basic principle of medication use during breastfeeding: ( )

A. Over-the-counter medications are safe to use during breastfeeding

B. If the medication used by the mother is also prescribed for newborns (infants), it is generally safe

C. When not affecting the therapeutic effect, choose medications with minimal transfer into breast milk and minimal impact on the newborn

D. For long-term or high-dose medication, monitor the infant's blood drug levels

B06. Colostrum is defined as the milk secreted within ( )

A. 5 days postpartum

B. 7 days postpartum

C. 10 days postpartum

D. Two weeks postpartum

B07. To increase milk production, a mother should pump every () hours on average, for more than ( ) minutes each time

A. 6; 15

B. 5; 30

C. 4; 30

D. 3; 15

B08. Which of the following statements about breast milk storage is incorrect: ( )

A. At room temperature 25-37°C, it can be stored for 4 hours

B. At room temperature 15-25°C, it can be stored for 8 hours

C. In the refrigerator at 2-4°C, it can be stored for 48 hours

D. In a freezer at -18°C or below, it can be stored for 3 months

B09. The best way to thaw breast milk is to take it out of the freezer and place it in ( )

A. Cold water

B. Bottle warmer

C. Refrigerator

D. Hot water

B10. Thawed breast milk should be warmed in water at ( ) °C:

A. 40-60 °C

B. 60-80 °C

C. 100 °C

D. 37-40 °C

B11. Thawed breast milk that has been heated but not consumed should be: ( )

A. Reheated for the next feeding

B. Discarded

C. Stored in the refrigerator

D. Stored in the freezer

B12. Which of the following statements is incorrect: ( )

A. The amount of collected breast milk should not exceed 3/4 of the container capacity

B. Breast milk can be stored in the refrigerator door

C. Use breast milk according to the collection time, prioritizing colostrum and fresh breast milk; for preterm and high-risk infants, fresh breast milk is best

D. During phototherapy, if continuous breastfeeding is required, avoid light exposure during feeding

Multiple-choice questions

B13. To protect, support, and promote

breastfeeding, WHO and UNICEF have formulated: ( )

A. “The Ten Steps to Successful Breastfeeding”

B. “The International Code of Marketing of Breast-milk Substitutes”

C. “The Hospital Breastfeeding Regulations”

D. “The Baby-Friendly Hospital Initiative”

B14. Measures to promote breastfeeding in hospitalized infants include: ( )

A. Informed choice

B. Establishing and maintaining lactation

C. Kangaroo care

D. Non-nutritive sucking

B15. The functions of colostrum include: ( )

A. Rich in antibodies

B. Acts as a laxative, promoting the passage of meconium

C. Contains growth factors, aiding in intestinal maturation

D. Rich in vitamins

B16. Which of the following is incorrect regarding medication use during breastfeeding: ( )

A. Estrogen in contraceptives affects the sexual development of infants and should be avoided

B. Taking thyroid hormone tablets should not be done while breastfeeding

C. Progestin-only contraceptives can be taken during breastfeeding

D. Mothers taking corticosteroids should stop breastfeeding

B17. When labeling breast milk sent to the hospital, in addition to the bed number and name, it should also include: ( )

A. Date

B. Time

C. Milk volume

D. Hospitalization number

B18. Methods to stimulate the let-down reflex include: ( )

A. Applying warm compresses to the breasts or taking a warm shower

B. Massaging the neck and back

C. Gently massaging, shaking, or tapping the breasts, and stimulating the nipple skin

D. Helping the mother relax and drink warm beverages

B19. To reduce contamination during breast milk collection, which of the following practices is correct: ( )

A. Both the mother and the assistant should wash their hands and disinfect the pump parts

B. Collecting naturally dripped breast milk is not recommended

C. It is not necessary to discard the first 5-10 ml of breast milk collected each time

D. Connect the collection parts directly to the storage container to reduce contamination

B20. Regarding breast milk transport, which of the following statements is correct: ( )

A. Maintain the cold chain state of breast milk: transportation temperature should be kept below 4°C

B. It is recommended to use cooling bags and dry ice for transport (for transport times over 18 hours, dry ice is recommended)

C. Regular ice can be used for insulating storage

D. Clean, dry towels can be used to fill the gaps between breast milk containers practices is correct.

Section 3: Questionnaire on Pediatric Medical-Surgical Nurses’ Attitudes Towards Breastfeeding for Hospitalized Newborns

Please answer the following questions based on your actual situation. In the answer area, tick “√” the option that matches your opinion. If you strongly agree or agree with a statement, tick “strongly agree” or “agree”. If you disagree or strongly disagree with a statement, tick “disagree” or “strongly disagree”. If you are unsure or do not know, please tick “not sure”.

No.	About Pediatric Medical-Surgical Nurses’ Attitudes Toward Breastfeeding Hospitalized Newborns	Strongly Agree	Agree	Not Sure	Disagree	Strongly Disagree
C-01	I believe that breastfeeding directly by					

	mothers is better than expressing milk for Pediatric Medical-Surgical babies					
C-02	I believe that breastfeeding is beneficial for Pediatric Medical-Surgical newborns					
C-03	I think mothers need more help from nurses to successfully breastfeed Pediatric Medical-Surgical babies					
C-04	I believe that the role of nurses is crucial in the breastfeeding of Pediatric Medical-Surgical babies					
C-05	I believe breastfeeding is better than formula feeding for Pediatric Medical-Surgical babies					
C-06	I am confident in my ability to support mothers in breastfeeding their Pediatric Medical-Surgical babies					
C-07	Our unit provides adequate support and resources for breastfeeding Pediatric Medical-Surgical babies					
C-08	I think more training on breastfeeding Pediatric Medical-Surgical babies is needed for nurses					

Section 4: Questionnaire on Pediatric Medical-Surgical Nurses' Practices of Breastfeeding for Hospitalized Newborns

The following items are a survey of Pediatric Medical-Surgical nurses' behaviors related to breastfeeding hospitalized newborns, based on your actual work situation. Please tick "√" the option that matches your actual work situation.

	No.	Pediatric Medical-Surgical Nurses' Breastfeeding Behaviors for Hospitalized Newborns	Never	Rarely	Sometimes	Often	Always
Breastfeeding Guidance	D-01	I provide handouts or brochures to families of Pediatric Medical-Surgical patients to monitor breast milk collection					
	D-02	I emphasize the importance of breastfeeding to the families of Pediatric Medical-Surgical patients					
	D-03	I actively provide knowledge about breastfeeding (such as teaching and demonstrating techniques, providing					

		supporting equipment, etc.), creating an encouraging environment to promote the initiation of breastfeeding					
Guidance on Collecting and Transporting Breast Milk	D-04	I guide families of Pediatric Medical-Surgical patients to collect breast milk within six hours after birth					
	D-05	I guide families to express milk 2-3 times per day					
	D-06	I guide families to wash their hands thoroughly before each breast milk collection					
	D-07	I guide families to clean the pumping room (without using disinfectants to avoid skin irritation)					
	D-08	I guide families on cleaning and disinfecting breast milk collection equipment					
	D-09	I guide families to store breast milk separately each time					
	D-10	I guide families to practice good hygiene when storing breast milk					
	D-11	I guide families to discard the first drops of naturally collected milk					
	D-12	I guide families to transport breast milk using coolers or dry ice, maintaining the cold chain during transportation					
Breast Milk Identification and Acceptance	D-13	I consult with physicians about medications taken by mothers to ensure they do not affect breastfeeding					
	D-14	I consult with physicians about mothers' illnesses to ensure they do not affect breastfeeding					
	D-	When accepting breast					

	15	milk, I check whether the labels are intact and clear (including date and time of collection)					
	D-16	When accepting breast milk, I check the volume and quality					
	D-17	When accepting breast milk, I sign according to the regulations					
	D-18	When accepting breast milk, I store it immediately for later use					
Storage, Thawing, and Heating of Breast Milk in the Ward	D-19	I check and clean the breast milk freezer in the ward every day					
	D-20	I separate and label each mother's breast milk in the freezer					
	D-21	I use breast milk according to the order of collection, prioritizing colostrum and fresh breast milk					
	D-22	I heat breast milk using a warm water bath					
	D-23	I freeze leftover breast milk after feeding					
Breastfeeding in the ward	D-24	When checking in new patients, I verify the identity and labels of the mother and baby					
	D-25	When checking in new patients, I check the milk for air bubbles					
	D-26	When checking in new patients, I check the color of the breast milk					
	D-27	During breastfeeding in the ward, I avoid exposing the milk to light					
	D-28	I pay attention to the feeding progress of babies in other wards as well					

Appendix 2

INFORMED CONSENT FORM

Project title: Knowledge, Attitude, and Behavior of Pediatric Medical Surgical Nurses on Breastfeeding: Basis for Capacity Building Plan

Purpose and conduct of the study

This study is an academic research project aiming to explore factors influencing the knowledge, attitudes, and practices of pediatric medical-surgical nurses regarding breastfeeding. By analyzing demographic characteristics and their relationship with breastfeeding-related behaviors, this research seeks to provide data-driven recommendations for capacity-building programs. All collected data will be used exclusively for academic purposes and will adhere strictly to ethical guidelines.

Participant's information (nature and extent of involvement)

You will be invited to participate in a 4-week cross-sectional study involving 150 pediatric medical-surgical nurses. If you decide to participate, you will be asked to answer a series of survey questions that will collect demographic information, including your age, sex, highest educational attainment, position category, years of nursing experience, years of pediatric medical-surgical experience, and the number of capacity-building activities you will have attended in the past year. Additionally, you will complete four questionnaires: the "General Information Questionnaire", the "Questionnaire on Breastfeeding Knowledge of Hospitalized Newborns," the "Questionnaire on Breastfeeding Attitude of Hospitalized Newborns," and the "Questionnaire on Breastfeeding Behavior of Hospitalized Newborns." The information collected from these questionnaires will help us understand your future knowledge, attitudes, and practices regarding breastfeeding and will serve as a basis for developing a capacity-building plan to enhance health promotion efforts in pediatric nursing.

Study-related treatments and probability for random assignment

In this study, proportional stratified random sampling will be used to ensure a representative sample of pediatric medical-surgical nurses from four anonymous hospitals in Shenzhen. A total of 253 nurses will be stratified by hospital, and the sample size from each hospital will be proportionally allocated based on the number of nurses. Specifically, Hospital A accounts for 23.7% of the total number of nurses, Hospital B 53.4%, Hospital C 13.0%, and Hospital D 9.9%. To achieve the final sample size of 150 participants, the sample from each hospital was adjusted to the nearest whole number: 36 nurses from Hospital A, 80 from Hospital B, 20 from Hospital C, and 14 from Hospital D. Minor rounding adjustments were made to ensure the total sample size of 150 while maintaining representativeness.

Within each hospital, simple random sampling will be applied to select participants from the eligible nurses. Each eligible nurse will be assigned a random number, and participants will be chosen using a random number table or computer-generated random numbers. This method ensures that all nurses have an equal probability of being selected, minimizing selection bias and ensuring a fair and representative sample.

To protect the anonymity of participants and smaller hospitals (e.g., Hospital D), all results will be aggregated during data analysis and reporting. Detailed demographic or professional information will not be disclosed in a way that allows identification of individual participants or hospitals. Additionally, specific results from smaller groups will not be reported independently, further safeguarding confidentiality.

Specific Procedures Based on Study Objectives

This study aims to explore the knowledge, attitudes, and practices (KAP) of pediatric medical-surgical nurses regarding breastfeeding. By analyzing the relationship between these factors and demographic characteristics such as age, education, and work experience, we seek to develop a capacity-building plan to enhance the support provided for breastfeeding in pediatric medical-surgical settings. The goal is to improve nurses' ability to support breastfeeding, thereby enhancing care quality and improving patient recovery outcomes.

Risk and inconveniences

There is virtually no risk involved in participating in this study, and no identifying information was collected. All data will be used solely for this study. On average, each questionnaire will take 20 minutes or less to complete. To minimize potential risks during face-to-face questionnaire completion, the process will be conducted in a private, well-ventilated environment to ensure your comfort and confidentiality. Hygiene measures, such as hand sanitization and optional mask usage, will also be implemented. Additionally, you may skip any questions that make you feel uncomfortable or withdraw from the study at any point without any consequences. To address any psychological discomfort, you will have access to contact information for support services if needed. The study team is committed to maintaining your privacy and confidentiality, ensuring that your responses cannot be traced back to you. If you have any questions regarding this study, you can contact Liu Liyue at +86 13631561198 or via email at liu.liyue@auf.edu.ph.

Possible benefits for the participants

As respondents in this study, there are no associated costs for participation. The study provides an incentive, offering respondents the chance to win a trophy through a lottery. This incentive is entirely optional and is not tied to the quality or completeness of participants' responses. It is solely a token of appreciation for the time and effort contributed by participants. The aim of the study is to assess pediatric medical-surgical nurses' knowledge, attitudes, and practices regarding breastfeeding.

The findings from this research can serve as a foundation for capacity-building programs, helping to enhance nurses' professional competencies in breastfeeding support. This, in turn, can improve maternal and infant health, promote better care practices, and reduce the burden on the healthcare system.

Contact persons

If you have any questions regarding the study or need assistance in answering the questionnaire, you may contact the corresponding author Ms. Liu Liyue at +86 13631561198 or via email at liu.liyue@auf.edu.ph.

Voluntariness of participation

Strict measures will be implemented to prevent any perceived coercion during the recruitment process. Recruitment will be conducted in collaboration with the hospital's nursing administration to identify eligible participants. However, you will be explicitly informed that your decision to participate, decline, or withdraw from the study at any time will not affect your employment, professional standing, or workplace relationships. All communication will emphasize the voluntary nature of the study, ensuring that you feel no obligation to participate due to your employer's involvement. Your participation in this research study is entirely voluntary, and you may choose not to participate or withdraw at any time without any obligations or consequences.

If any new information arises during the study that could affect participants' willingness to continue, participants or their legally acceptable representatives will be informed promptly. Should a participant choose to withdraw from the study, all associated data will be destroyed and will remain confidential and anonymous.

Confidentiality and data management

Before each respondent registers for this study, the researchers will be responsible for providing a comprehensive and detailed introduction to the study's purpose, procedures, and potential risks, and obtaining a signed written informed consent form. To ensure anonymity and blinding in this study, all participant data will be anonymized by assigning unique codes. Personal identifiers such as name, hospital will not be included in the dataset used for analysis. Consent forms will be stored separately from the survey data to prevent identity linkage. The researcher will only analyze de-identified data, and any data issues or anomalies will be addressed without linking back to individual participants' identities. Data analysis and reporting will be conducted in an aggregated format, ensuring no individual participant can be identified. Respondents will have the right to withdraw from the study at any time. Upon withdrawal, their personal information and data will be destroyed and will not be disclosed to any third party. The personal privacy and data confidentiality of respondents will be protected during the study.

After respondents complete the questionnaire, the research data will be collected and managed by the researchers to ensure the accuracy of the experimental data and the privacy and confidentiality of respondents. The original questionnaires will be kept by the researchers and stored in a separate locked cabinet. Respondents will be coded, and the data will be analyzed in a blinded manner, with unresolved issues in the data being reviewed. The stored data will not include the names of respondents, meaning that the published data analysis cannot be traced back to the respondents. All other relevant information, including answers to questions in the survey, will be used solely for research purposes. The actual survey forms and summary tables will be kept for three years after the study concludes, after which they will be destroyed using a shredder, and a final check will be conducted to ensure that all data and questionnaires in the drawer have been completely destroyed. The results of all questionnaires will not be used directly or indirectly for any other research.

The data management practices of this study fully comply with all relevant national and international data protection regulations, including China's Personal Information Protection Law (PIPL) and the General Data Protection Regulation (GDPR). These measures ensure that participants' data are handled securely and ethically, meeting the highest standards of privacy protection.

Access to the results of the study

The participants shall be immediately notified once the study is done. If the participants choose to be informed of the results, the researchers will be able to do so upon submission of a request to the corresponding author. To ensure effective communication of the study results, the researchers will implement a detailed dissemination plan. Results will be shared in an accessible format, such as summary reports or visual presentations, within three months after the study concludes. Participants may request the results through the corresponding author, and these will be provided in a clear, non-technical language that ensures all participants can easily understand the findings.

Only the authors, participants, and those who are directly concerned with the study (e.g., Ethics Review Committee, etc.) have the right to access the results.

Withdrawal Criteria

Participants may withdraw from the study at any time without any obligations or consequences. In the event of unforeseen interruptions, such as natural disasters, public health emergencies, or institutional changes, the researcher will notify the Ethics Committee and provide plans for restarting or halting the study.

Participation in this study may also be terminated under the following circumstances:

- Non-compliance with study procedures that compromises data integrity.
- Significant health-related changes or adverse events that make participation unsuitable.
- Regulatory or institutional requirements that necessitate study termination.

Participants will be informed promptly of any termination reasons and follow-up actions. All data from withdrawn or terminated participants will be excluded from analysis and securely destroyed to maintain privacy and confidentiality.

Ethical approval

This study has been approved by the Ethics Review Committee of Suining Central Hospital as a research site. For clarification, please contact the Ethics Committee at 0825-2292068. The Ethics Review Committee of Angeles University Foundation has approved this study. If you have any questions, the committee can be reached through ERC chair:

Dr. Bella G. Panlilio

Chair, ERC

Ethics Review Committee, Angeles University Foundation

panliliobg@auf.edu.ph

(045) 625 2888 local 170

As part of compliance with ethical and regulatory standards, study monitor(s), auditor(s), the AUF-ERC, and relevant regulatory authorities will be granted direct access to participants' medical records. This access will strictly adhere to confidentiality and data protection regulations, ensuring that participants' privacy and anonymity are preserved throughout the review process.

Statement of the Participant

I have read and understood the above information and have been given the opportunity to consider and ask questions on the information regarding my involvement in this study. I have spoken directly to the investigator/s of this study who have answered to my satisfaction all my questions. I have received a copy of this Participant's Information and Informed Consent Form. I hereby voluntarily agree to participate.

Name of Participant	Signature of Participant	Date

Statement of the Researcher

I hereby attest that the participant has read and understood the above information and has been given the opportunity to consider and ask questions on the information regarding the involvement in this study. The participants were also given a copy of this Participant's Information and Informed Consent Form. I also attest that the participant has volunteered for this study and was not coerced.

Name of Participant	Signature of Participant	Date

Appendix 3 Gantt Chart