Nanoparticle technologies to enrich aqueous solutions with oxygen for healing applications

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Purpose

This systematic analysis aims to:

- categorize the dissolved oxygen (DO) technologies tested in healing protocols to date, based on the properties of the oxygen-generating nanoparticles used in each approach
- investigate their outcomes on wound oxygenation and healing efficacy
- followed by a critical overview of the biological and administration parameters that require further evaluation

Introduction

- Non-healing wounds represent an emerging global-health issue, given the prevalence of predisposing comorbidities such as diabetes.
- Oxygen (O_2) is known to play a fundamental role in all phases of the healing physiology, while prolonged hypoxia predisposes to wound chronicity.
- DO solutions might introduce an innovative approach to confronting chronic wounds;
  - in theory, such formulations could combine the established benefits of an aqueous environment, with the enhanced oxygenation of the wound bed that nanoparticle technologies enable.

Methodology

Systematic review, based on PRISMA statement and PICOS framework

- Formulations including artificial oxygen carriers were excluded from the analysis, to avoid potentially confounding effects
- All clinical and experimental studies evaluating DO technologies in healing were included

- For each healing protocol, the following parameters were analyzed: level of evidence, subject, number of wounds/subject, wound model, duration and frequency of intervention(s), treatment group(s), control group(s)
- Nanoparticle technologies generating DO solutions were evaluated based on the following criteria: manufacturing and application details; O_2 supply/origin during manufacturing, additional materials (apart from O_2), resulting O_2 levels, physical parameters.

Results

Our search yielded 20 original articles, categorized in 4 technological approaches (**Table 1, Figure 1**):

- DO solutions containing O_2 nanoparticles (ONPs)
- DO solutions containing O_2 nanobubbles (ONBs)
- Super-saturated O_2 solutions (SOSs)
- O_2 emulsions

DO technology	Engineering approach	Significant increase in O_2 levels (within 3h from production)	Maximum duration of oxygenating effect	First author, Year
ONPs	High-speed centrifugation to produce nanoparticles; homogenization with exogenous core and/or coating materials with enhanced O_2-binding capacity; O_2 delivery enhanced with ultrasound energy	DO = 0.016 - 4.5mg/mL	*in vitro*: 24h *in vivo*: 15min	Magnetto et al. 2014; Khadjavi et al. 2015; Pratto et al. 2015; Gulino et al. 2015; Basilico et al. 2015; Bisazza et al. 2008; Cavalli et al. 2009 (a); Cavalli et al. 2009 (b); Feshitan et al. 2014; Fiala et al. 2020
ONBs	3 steps: 1) hydrodynamic mixture of O_2 with normal saline to produce O_2 microbubbles; 2) ultrasound for 30s; 3) new hydrodynamic mixture with O_2 application to produce O_2 nanobubbles	PO_2 = 1003.2 - 1053.3mmHg	N/A (mixed with hypoxic normal saline)	Matsuki et al. 2012, 2014
		DO = 0.018 - 0.032mg/mL	*in vitro* & *in vivo*: 70 days	Ebina et al. 2013; Noguchi et al. 2017; Matsuoka et al. 2018
SOSs	Oxygenated saline solution; enriched with oxygen and chloride free radicals via electrolysis	DO = 0.009 - 4.0mg/mL	N/A (administered daily to maintain effect)	Onouye et al. 2000; Paola et al. 2006
O_2 emulsions	Emulsion in cream format by applying a surfactant oil in a solution containing O_2 and perfluorocarbon nanoparticles; 20 times higher O_2 solubility than water	2ml O_2 per ml of emulsion	N/A	Davis et al. 2007
		T_cPO_2 = 22 - 25mmHg	7 days	Li J et al. 2013, 2015

Table 1: Engineering and oxygenating parameters of the DO technologies from our analysis.

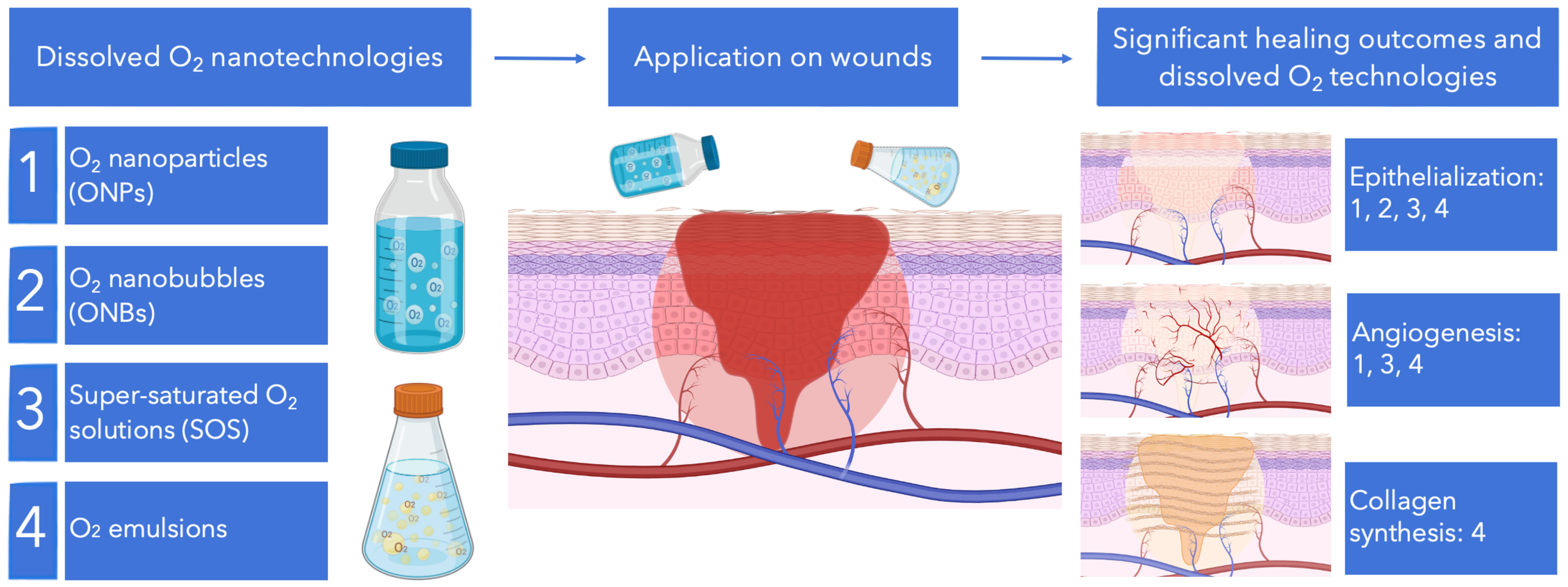

Figure 1: Statistically significant healing outcomes per DO technological group from our analysis.

Discussion

- ONBs might represent the most promising technological approach to producing DO solutions; their nanoscale size was accompanied by significant physical stability, enabling consistent O_2 supply for up to 70 days.
- Exogenous materials such as perfluorocarbons and surfactant oils raise multifaceted safety concerns *in vivo*, due to reports of significant and prolonged toxicity to vital organs.
- Statistical comparisons among the DO technological approaches were impeded by the significant methodological variability, in both the healing and the oxygenation endpoints.

Conclusions

- Among DO technologies, ONB solutions may establish the O_2 levels required for uncomplicated healing for the longest duration, with the least safety concerns.
- Targeted experiments comparing all DO technologies in the same healing protocol and with a standardized methodology are highly encouraged; this would be a key step before testing them in the rapidly-expanding cohort of non-healing wounds globally.

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