



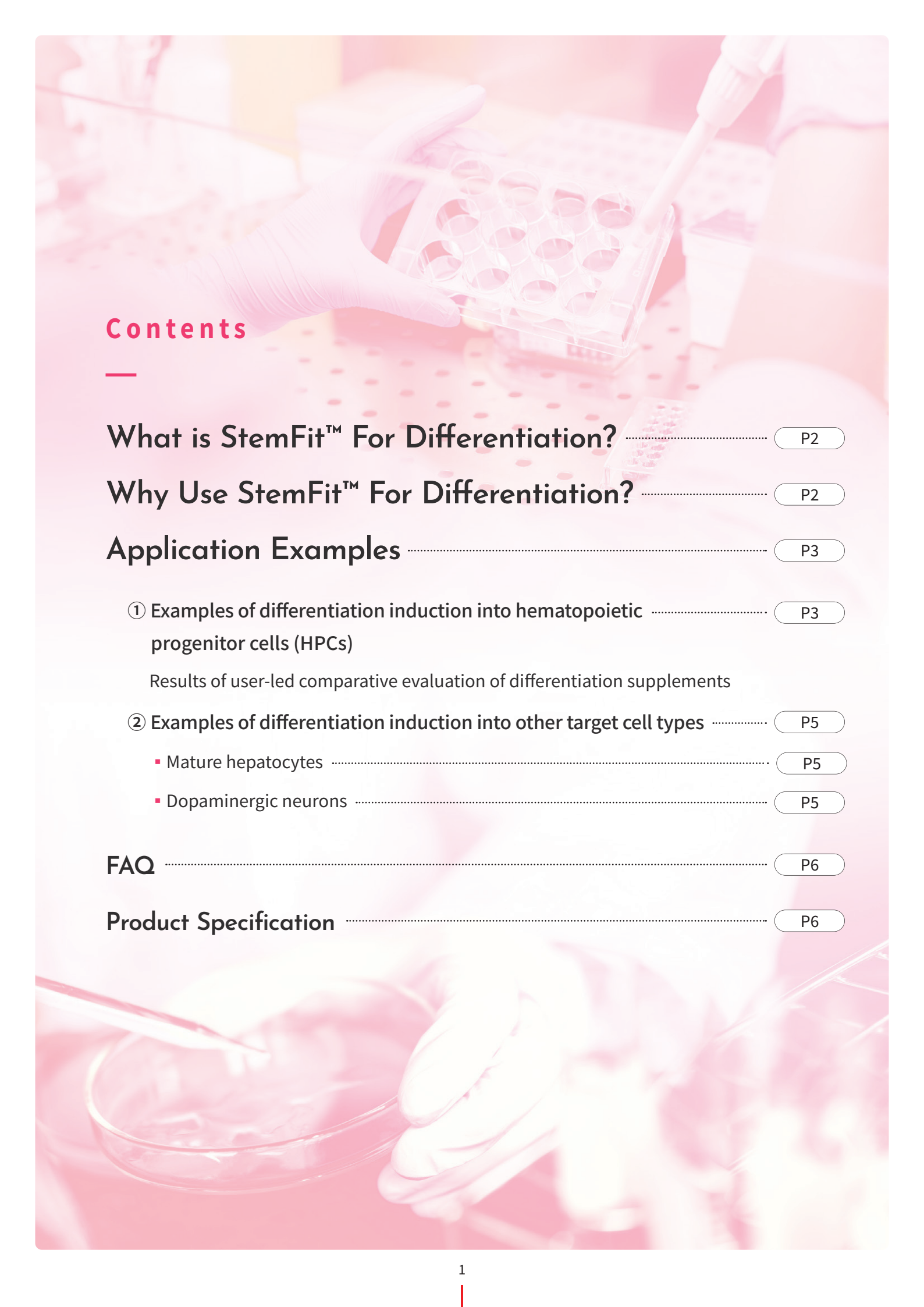
StemFit™

For Differentiation



From Bench to Clinic: Serum-free Differentiation

- ✓ Consistent High Performance and Reproducibility
- ✓ Clinically Applicable
- ✓ Reduced Risk of Contamination



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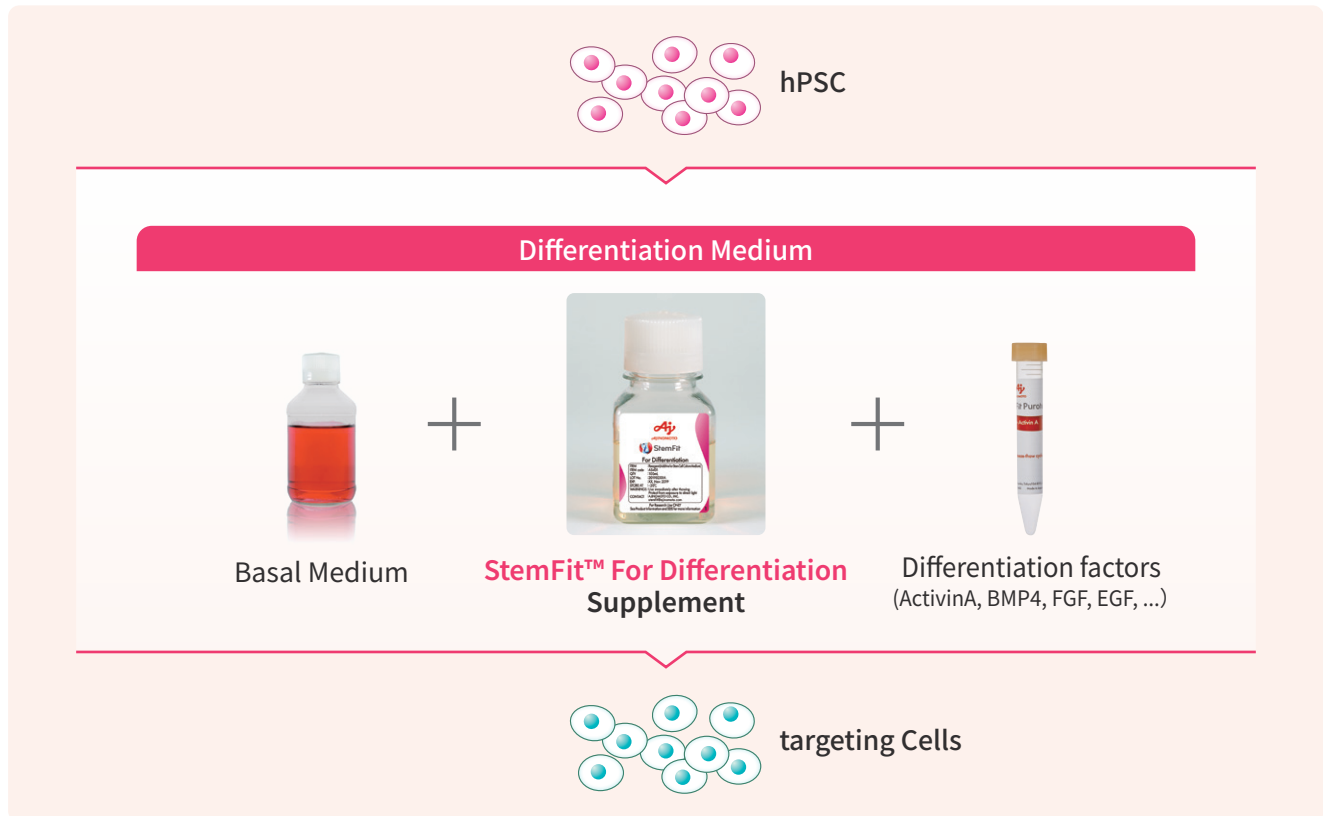
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What is StemFit™ For Differentiation?

StemFit™ For Differentiation (Diff.) is a defined, animal-origin-free (D-AOF) supplement designed for the differentiation of human ES and iPS cells. StemFit™ For Diff. is provided as a concentrated solution (containing albumin) and is intended to be used in combination with basal media such as DMEM/F-12, RPMI, or DMEM, together with various differentiation factors and cytokines, including Activin A, BMPs, FGFs, and EGF.



Why Use StemFit™ For Differentiation?

High Performance and Reproducibility

- Achieves **high differentiation efficiency and yield** across multiple hiPSC lines
- Defined composition and AOF **minimize lot-to-lot variability**

Reduced risk of Contamination

- **Eliminates the risk** of animal-derived viruses and undefined components
- Provides a serum-free clean culture environment

Clinically Applicable

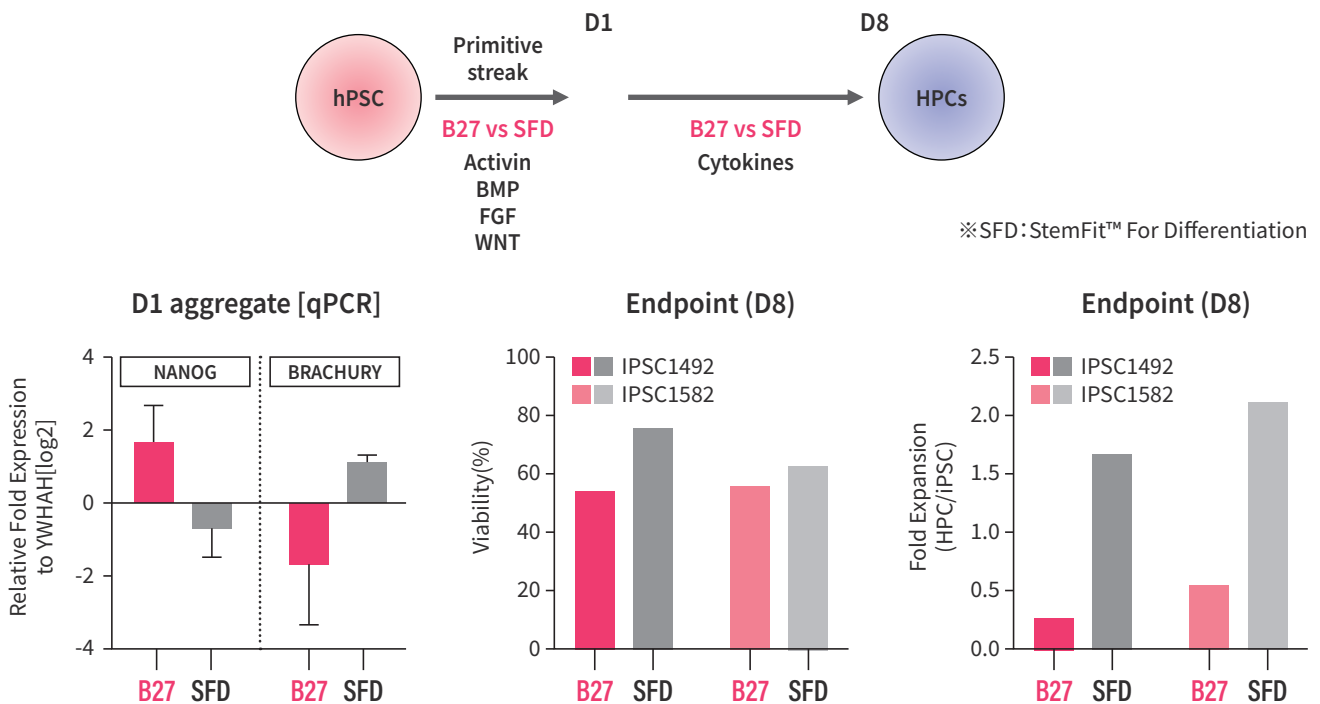
- Defined AOF **facilitates regulatory submissions and compliance**
- Produced under **GMP standards**, supporting clinical applications (CoA, CoO, GMP Statement Available)



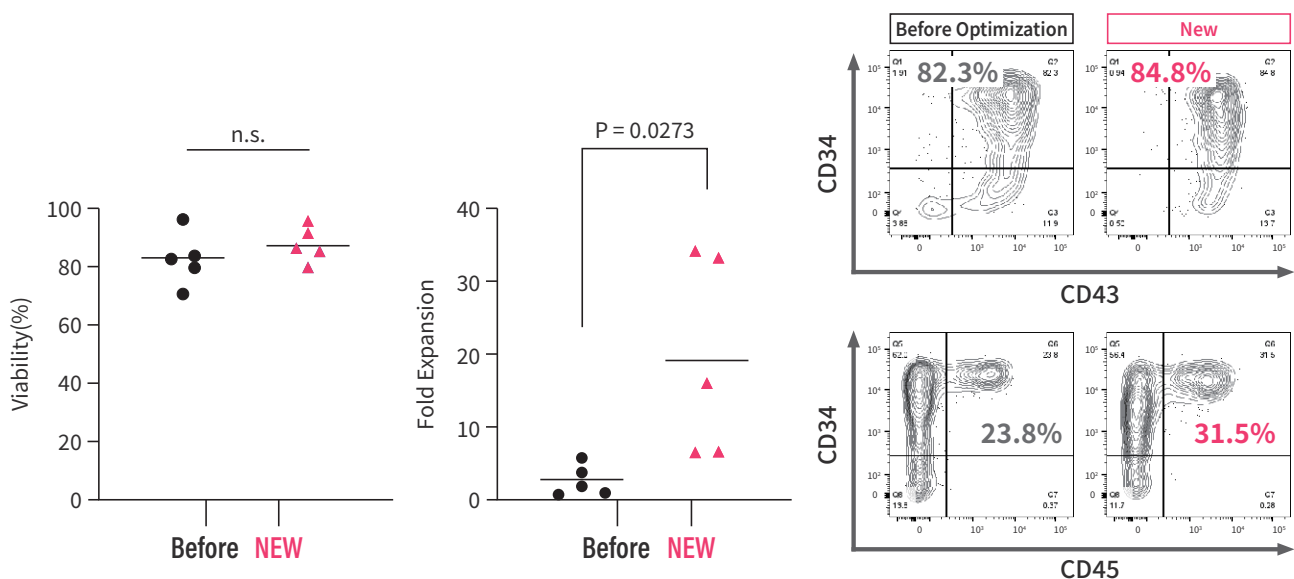
Application Examples

1 Example of differentiation induction into hematopoietic progenitor cells (HPCs) – Results of a user-led comparative evaluation of differentiation supplements

Century Therapeutics, Inc. evaluated iPS-to-HPC differentiation using StemFit™ For Differentiation with the aim of improving T-cell differentiation efficiency. Using an 8-day differentiation process, they confirmed a consistent increase in HPC yield as well as enhanced differentiation efficiency toward lymphoid lineages.

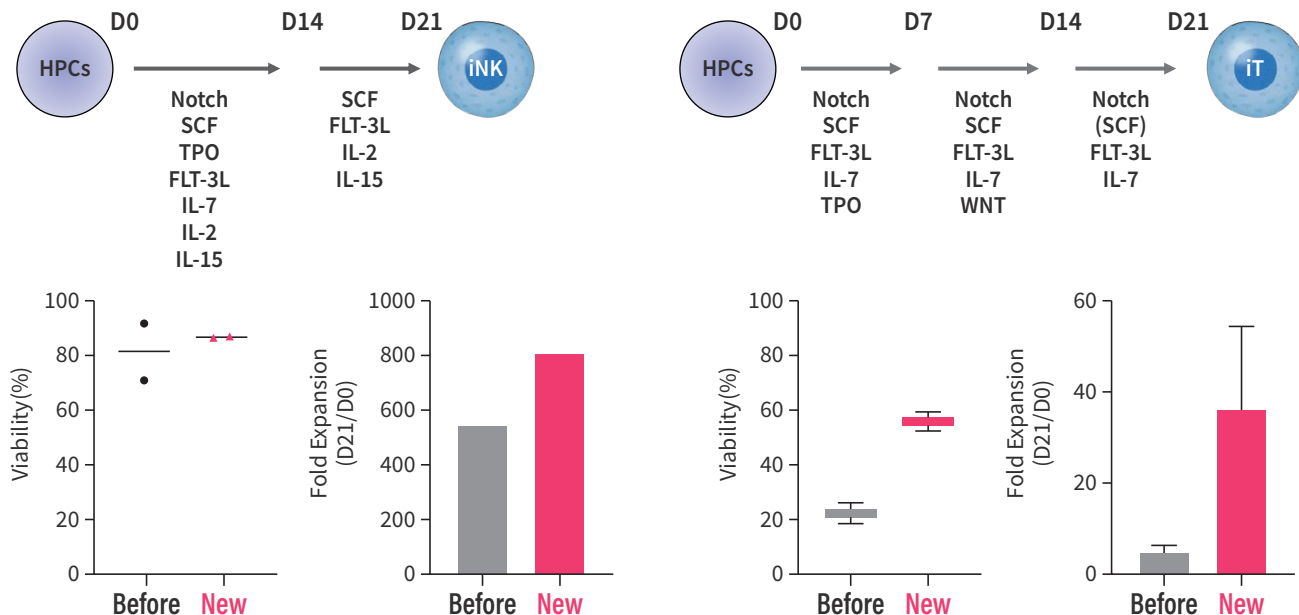


Furthermore, following protocol optimization using StemFit™ For Differentiation, a substantial improvement in HPC yield was observed while maintaining cell viability and the proportion of CD34+CD43+ cells. This trend was consistently reproduced across multiple iPSC lines, including PiPSCs and TiPSCs, and among multiple operators with varying levels of experience.

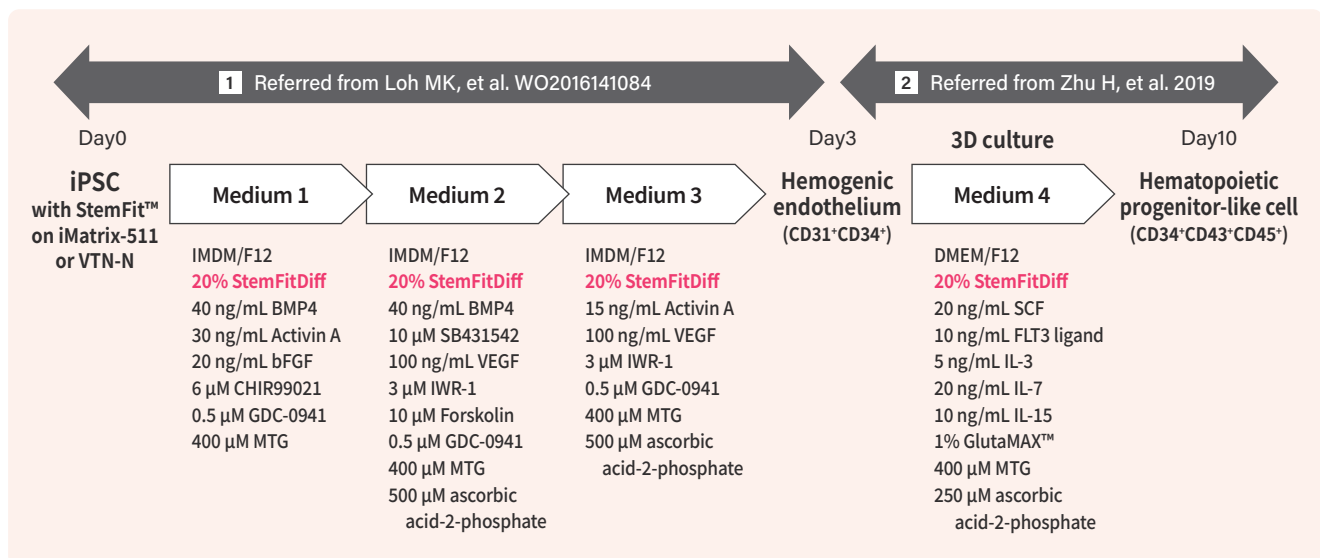


► Example of differentiation induction into lymphoid cells (iNK / iT) via HPCs

Using the obtained HPCs, further differentiation toward lymphoid lineages was evaluated. These results demonstrate differentiation from HPCs to iNK and iT cells using StemFit™ For Differentiation. Following protocol optimization, an improvement in differentiation efficiency toward lymphoid lineages was confirmed.



► Example protocol for differentiation from iPS cells to hematopoietic progenitor cells (HPCs)



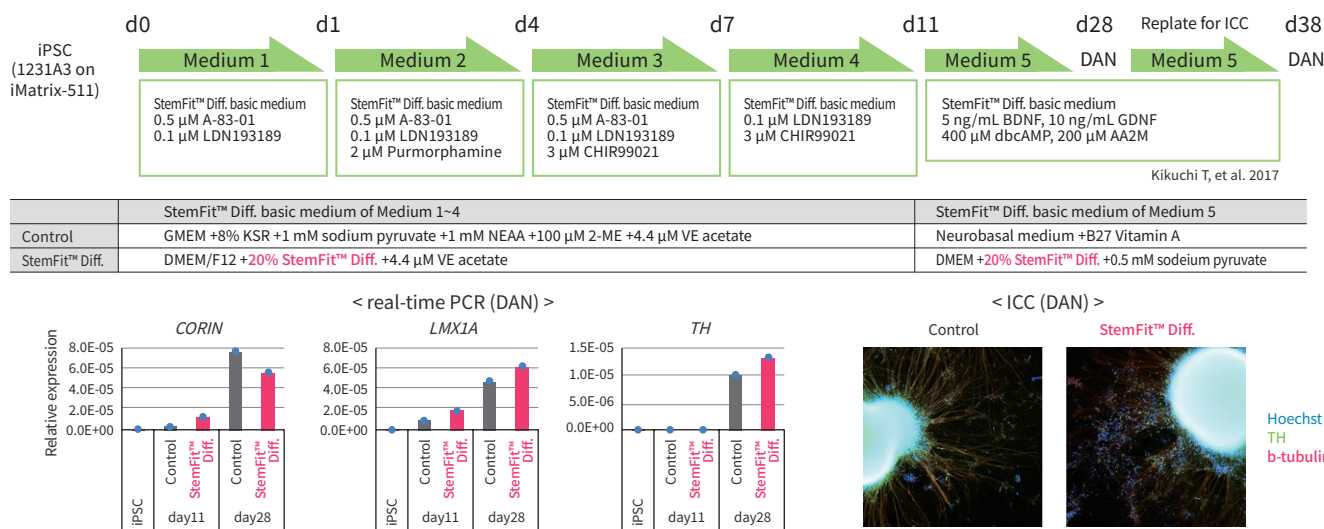
Partially-confluent wells of undifferentiated hiPSCs were dissociated into single cells using Accutase and sparsely passaged on 0.5 μg/cm² iMatrix-511 or VTN-N coated 6 well plate at 1.0 – 3.0 x 10⁵ cells in StemFit™ Basic03 supplemented with 80 ng/mL bFGF and 10 μM Y-27632 overnight. Seeding hiPSCs sparsely prior to differentiation was critical to prevent cellular overgrowth during differentiation, especially during longduration differentiation. hiPSCs were allowed to plate overnight, and the following morning, were briefly washed (in IMDM/F-12) and differentiated towards Primitive Streak (PS) using Medium 1 for 24 hours. Day 1 PS was subsequently washed (IMDM/F-12) and further differentiated using Medium 2 for 24 hours. Day 2 cultures were further directed into HE by Medium 3 for 24 hours.

2 Examples of differentiation induction into other target cell types

StemFit™ For Differentiation is a highly versatile supplement that supports differentiation of iPS cells into all three germ layer lineages. To date, differentiation has been confirmed toward mesodermal lineages (e.g., HPCs and macrophages), ectodermal lineages (e.g., dopaminergic neurons), and endodermal lineages (e.g., hepatocytes and renal cells). The protocols shown below represent selected examples of these differentiation applications.

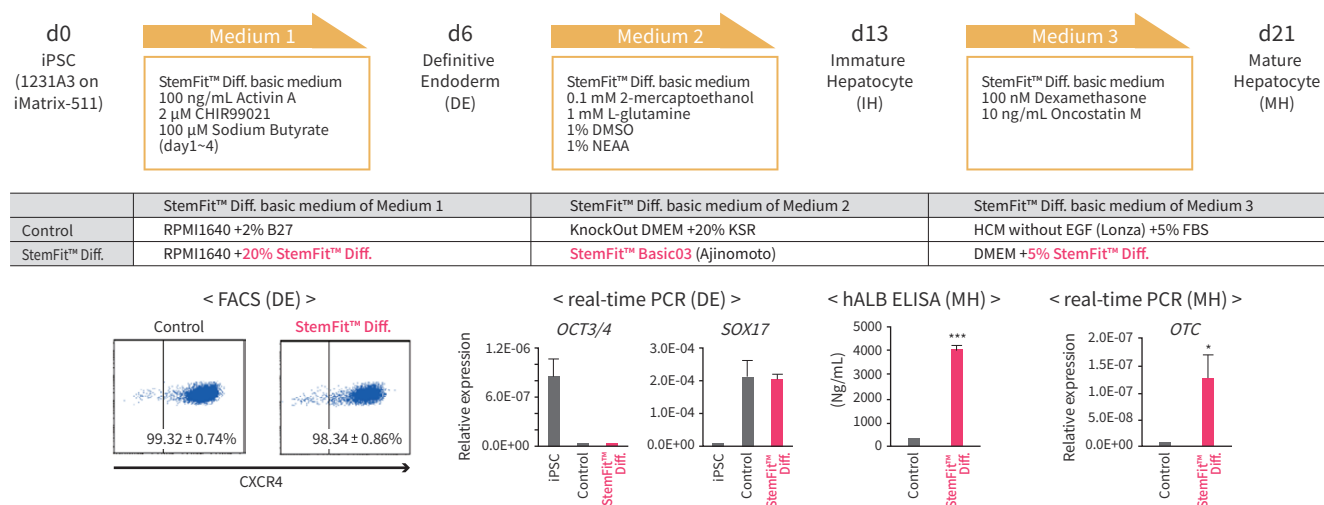
iPS to Dopaminergic Neuron

Superior differentiation performance was observed when **KSR** or **B27** was replaced with StemFit™ For Differentiation.



iPS to Hepatocyte

Superior differentiation performance was observed when **FBS**, **KSR**, or **B27** was replaced with StemFit™ For Differentiation.



FAQ

Q Which basal medium can be used with StemFit™ For Differentiation (StemFit™ For Diff)?

StemFit™ For Diff. can be used with any Basal medium (DMEM, RPIM1640, DMEM/F12) which are used for iPS/ES cell differentiation

Q What is the recommended final concentration of Stemfit™ For Diff.?

We usually recommend final concentration of 20%. For example, mix 100 ml of StemFit™ For Diff. with 400 ml of basal medium to prepare differentiation basic medium. Lineage specific cytokines or small molecules should be added to differentiation basic medium.

Q Can I use StemFit™ For Diff. for all lineage?

We have confirmed that StemFit™ For Diff. can be used for EB formation and directed differentiation for endodermal, mesodermal and ectodermal lineages. Please refer the [product information](#). There are some cases where protocol optimization would be required. Please contact us directly for more information.

Q Does it contain insulin?

Yes, it does.

Q Are there any target cell types that are difficult to differentiate using StemFit™ For Differentiation?

There are cases in which differentiation into cardiomyocytes may be challenging.

Q Are there other applications?

Please refer to the collection of publications. [Click here](#) to view the publication list.

Product Specification

Product Name	StemFit™ For Differentiation
Product Number	SF051-001
Volume	100ml
Storage	Store at -20±5°C

StemFit™ For Differentiation is for research and further manufacturing use only and is not intended for Human or Animal diagnostic or therapeutic uses.

GMP-Compliant Quality System

The Quality Management System (QMS) of this product is in compliance with Ajinomoto's quality assurance framework, ASQUA (Ajinomoto System of Quality Assurance). Manufacturing and product release testing are managed based on GMP standards established by Ajinomoto specifically for regenerative medicine media. These GMP standards have been developed in accordance with ICH Q7 and the PMDA Guidance on Aseptic Manufacturing of Regenerative Medical Products.

Please click the URL or scan QR code to see more details:

<https://www.ajittrade.com/stemfit/GMP-guideline/>



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Eat Well, Live Well.



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