Particle assisted dermal penetration – a mechanistic proof of concept study

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Supplementary material – Section S1

All images were subjected to an automated RGB-threshold algorithm. The macros were established with ImageJ software for each part of the study to adjust for the variations in the skin`s autofluorescence. The macros used are provided in table S1.

Table S1: Macros used for the RGB-threshold algorithms.

Particle assisted dermal penetration with titanium dioxide particles	Particle assisted dermal penetration with NLC	Particle assisted penetration and addition of humectants

// Color Thresholder 1.52a
// Autogenerated macro, single images only!
min=newArray(3);
max=newArray(3);
filter=newArray(3);
a=getTitle();
run("RGB Stack");
run("Convert Stack to Images");
selectWindow("Red");
rename("0");
selectWindow("Green");
rename("1");
selectWindow("Blue");
rename("2");
min[0]=0;
max[0]=0;
filter[0]="stop";
min[1]=100;
max[1]=255;
filter[1]="pass";
min[2]=0;
max[2]=0;
filter[2]="stop";
for (i=0;i<3;i++){
selectWindow(""+i);
setThreshold(min[i], max[i]);
run("Convert to Mask");
if (filter[i]=="stop") run("Invert");
	// Color Thresholder 1.52a
	// Autogenerated macro, single images only!
	min=newArray(3);
	max=newArray(3);
	filter=newArray(3);
	a=getTitle();
	run("RGB Stack");
	run("Convert Stack to Images");
	selectWindow("Red");
	rename("0");
	selectWindow("Green");
	rename("1");
	selectWindow("Blue");
	rename("2");
	min[0]=0;
	max[0]=0;
	filter[0]="stop";
	min[1]=60;
	max[1]=255;
	filter[1]="pass";
	min[2]=0;
	max[2]=0;
	filter[2]="stop";
	for (i=0;i<3;i++){
	selectWindow(""+i);
	setThreshold(min[i], max[i]);
	run("Convert to Mask");
	if (filter[i]=="stop") run("Invert");
		// Color Thresholder 1.52a
		// Autogenerated macro, single images only!
		min=newArray(3);
		max=newArray(3);
		filter=newArray(3);
		a=getTitle();
		run("RGB Stack");
		run("Convert Stack to Images");
		selectWindow("Red");
		rename("0");
		selectWindow("Green");
		rename("1");
		selectWindow("Blue");
		rename("2");
		min[0]=0;
		max[0]=0;
		filter[0]="stop";
		min[1]=119;
		max[1]=255;
		filter[1]="pass";
		min[2]=0;
		max[2]=0;
		filter[2]="stop";
		for (i=0;i<3;i++){
		selectWindow(""+i);
		setThreshold(min[i], max[i]);
		run("Convert to Mask");
		if (filter[i]=="stop") run("Invert");

```
}
imageCalculator("AND create", "0","1");
imageCalculator("AND create", "Result of 0","2");
for (i=0;i<3;i++){
   selectWindow(""+i);
   close();
}
selectWindow("Result of 0");
close();
selectWindow("Result of Result of 0");
rename(a);
// Colour Thresholding----------
run("Invert");
```


```
}
imageCalculator("AND create", "0","1");
imageCalculator("AND create", "Result of 0","2");
for (i=0;i<3;i++){
   selectWindow(""+i);
   close();
}
selectWindow("Result of 0");
close();
selectWindow("Result of Result of 0");
rename(a);
// Colour Thresholding----------
run("Invert");
```

```
}
imageCalculator("AND create", "0","1");
imageCalculator("AND create", "Result of 0","2");
for (i=0;i<3;i++){
   selectWindow(""+i);
   close();
}
selectWindow("Result of 0");
close();
selectWindow("Result of Result of 0");
rename(a);
// Colour Thresholding----------
run("Invert");
```