## Skin micro-channels created by Spongilla spicules

SHS can insert into skin with a depth of  $48.6 \pm 13.5 \mu m$  (n=100) and with a distribution of  $850 \pm 125$  spicules per mm2 (n = 6) in the treatment area after SHS treatment immediately. (10mg per 1.77cm2 with electric massage for 2min) (Zhang et al. 2019)



Figure 1 The distribution profile of the potential microchannels created by SHS in skin over time in vivo. There were 850 ± 125 spicules per mm2 (n = 6) on guinea pig skin topical applied by SHS application (10mg per 1.77cm2 with electric massage for 2min)

- The surface area of adults is about 18,000 cm2 (men) or 16,000 cm2 (women).
- The face is 2% of the total surface area, : 320 cm2
- 320cm2 = 32000 mm2
- 850 microchannels x 32000mm2 = 27 200 000 microchannel
- 85 microchannels x 32 000mm2 = 2.3 mil microchannels (in our professional treatment – 3g (0.47 mg spicules/cm).

## Our Bio needle professional dose

We use 5% of the 99% spicules in Formulation

We use 3g of products per treatment which equates to 150 mg of spicules which would be 0.47 mg/cm or 0.83 mg/1.77cm2 (to directly compare to article).

Our product is 12 x less concentrated than the article, thus the number of microchannels will be theoretically 12 x less (in 2 minute massage)

We do massage for longer which would cause more channels but this cannot be quantified and the number of micro channels cannot be assumed to be proportional to massage duration.

## References

Adapted from Zhang, C., Zhang, K., Zhang, J., Ou, H., Duan, J., Zhang, S., Wang, D., Mitragotri, S. and Chen, M., 2019. Skin delivery of hyaluronic acid by the combined use of sponge spicules and flexible liposomes. Biomaterials science, 7(4), pp.1299-1310.