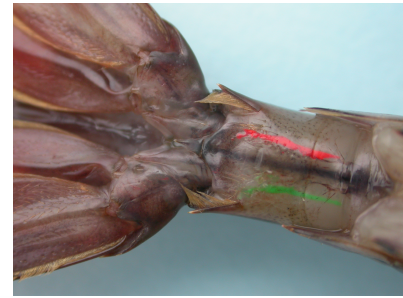



Northwest Marine Technologies, Inc. Visible Implant Elastomer Tags



Visible Implant Elastomer is injected as a liquid and remains visible through clear or transparent tissue.

 **Visible Implant Elastomer (VIE) Tags** are colored fish tags. They are also used to tag crustaceans, cephalopods, gastropods, reptiles, amphibians, small mammals, arachnids, and other animals. The elastomer is injected as a liquid that cures into a pliable solid. The tags are implanted beneath clear or translucent tissue and remain externally visible. Visible Implant Elastomer tags are small, flexible, and bio-compatible. This makes them useful for a wide variety of species and in animals smaller than most other tags can accommodate.

Visible Implant Elastomer, or VIE, was first developed in the 1990's as a fish tag to provide a visible, internal tag to avoid some of the negative effects associated with external tags. Because it has some unique properties, VIE has turned out to be very versatile for animal tagging. It has been applied to a wide range of sizes and species, both on land and in the water. It has been used on all continents, and in all sorts of environments. There is a large body of publications and reports about using VIE, and below are some examples that demonstrate it's versatility. Please contact NMT if we can help you find publications about a species you may like to tag.

Fish

VIE was first used as a fish tag, and this remains the most popular application. Species representing at least 65 families have been tagged with VIE all over the world to help us understand their behavior, biology, ecology, and management. Zebrafish (*Danio rerio*) are commonly used to study human diseases, and these little fish can be identified with VIE (Hohn & Petrie-Hanson, 2013). Another unusual use of VIE with fish is for tagging small sharks (Jacoby et al 2014).



Cephalopods

Octopus are admired for their ability to crawl in and out of tight spaces, but did you know that they are also quite adept at removing external tags? VIE has provided a nice solution for tagging octopus, squid and cuttlefish. The tags are quickly injected without anesthetic, remain highly visible and are suited for individual identification by combining tag locations and colors (Brewer & Norcross, 2012; Replinger & Wood, 2007; Sykes et al. 2012; Sykes et al 2017)



Photo by J. Wood

Echinoderms

While fascinating to study, echinoderms are notoriously difficult to tag. There have been a few successes, and Martinez et al (2013 ; 2018) overcame the dark, thick skin that often obscures VIE tags in echinoderms by adapting a very powerful underwater light to fluoresce the tags in their studies of seastars.

Slugs and Worms

While not related to each other, the application of VIE to these crawlers is similar, and has provided a neat solution to the problems of tagging animals whose skin is continually in contact with abrasives. To our knowledge, Butt & Lowe (2007) were the first to apply VIE to annelids, followed by Wallin & Latty (2008) in applying it to slugs.

Amphibians and Reptiles

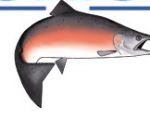
VIE offers a humane and practical alternative to toe clipping for identifying amphibians and reptiles, and has been widely used in a number of species. A novel application for amphibians was its use in identifying salamander egg masses (Regester & Woosley 2005). Among the reptiles tagged with VIE are lizards, snakes (Hutchens et al 2008), and turtles. Hatchling turtles have been tagged in two ways. Davy et al. (2010, see volume 41(4)) injected the tags into the limbs, while Anderson et al (2015) adapted a binary coding scheme to the plastron to create individual tag numbers.

Arachnids

Tagging tarantulas is not for everyone, but Melo et al (2013) showed that it can be done with VIE. Similarly, Chapin (2011) found VIE to be a viable method of tagging scorpions.

Small mammals

Visible Implant Elastomer has been used to identify shrews and other small mammals, generally by placing the tags at the base of the tail. See theses by Rentz 2014 and Smith 2012 for examples.



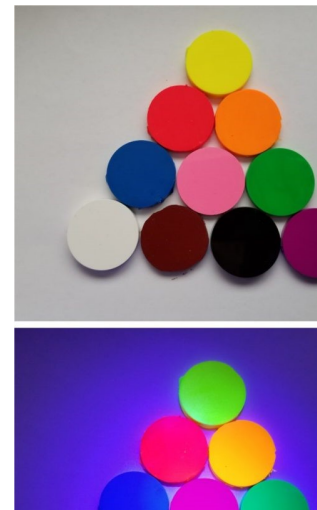
Color Selection

Visible Implant Elastomer comes in 10 colors. Red, pink, yellow, green, orange and blue are fluorescent. Black, purple, brown, and white are not. The fluorescent colors are highly visible in ambient light and provide the option of greatly enhanced tag detection when fluoresced with the VI Light. Fluorescing the tags is especially helpful when tagging under pigmentation or when recovering tags in the dark.

VIE color selection can be tricky and getting it right is an important part of your experimental design. Your choice depends on how much contrast you need with the background pigmentation, how many different colors you require, and the type of light you will use to fluoresce the tags.

General recommendations

- ▶ Exhaust all possibilities with fluorescent colors before using non-fluorescent colors. Maximum tag detection is obtained by fluorescing tags with the VI Light.
- ▶ Use non-fluorescent colors only when there is no potential for the tag to become obscured and when all tag detection will occur in bright light.
- ▶ When using the VI Light, we recommend that the first four colors selected be red, yellow, orange, and blue.
- ▶ Test the product under field conditions before beginning large-scale projects.
- ▶ Colors that may be confused when using the VI Light:
 - ▶ Green and yellow appear nearly identical.
 - ▶ Red and pink look similar but can be separated if the VIE Color Standard is used and if the samplers are trained. We recommend that you test these two colors in your animal and with the samplers who will recover the tags.
- ▶ Colors that may be confused in Ambient Light:
 - Black and brown appear nearly identical.
 - Brown and purple look similar. With the VI Light, purple will appear much lighter while brown does not change. This feature may be used to help distinguish these colors.



Visible Implant Elastomer is available in 10 colors, shown on the top, in ambient light. Red, green, yellow, orange, blue, pink and green fluoresce under the VI Light (bottom photo).

Generate a Coding Scheme

While VIE is primarily used for batch identification, you can generate a VIE coding scheme by combining multiple tags, tag locations, and colors. For example, researchers tracking seahorses used this method to distinguish more than 500 individual seahorses at one time.

Six fluorescent colors (red, yellow, blue, orange, green, and pink) and four non-fluorescent colors (black, brown, white, and purple) constitute the only intrinsic VIE codes. If you use just one tag per fish, the number of unique codes is simply the product of the numbers of colors used and the number of tag locations.

However, if you inject two or more tags in each animal, in combination with several different locations and colors, you can generate an extensive VIE coding scheme. In any tagging program, it is important that all of the study animals have the same number of tags. That way, there can be no confusion between animals which lost tags and those which began with fewer tags.

How many codes?

Use NMT's app to generate a VIE coding scheme. You can download a free copy from www.nmt.us

Calculate the number of unique codes you could create using the formula:

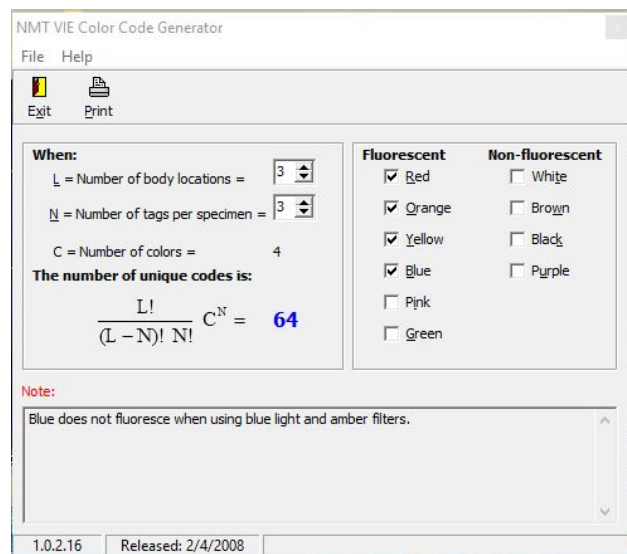
$$\frac{L!}{(L-N)! N!} C^N$$

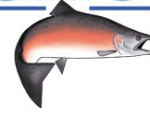
Where: C= Number of colors used, L= Number of body locations and N = Number of tags per animal.

For example, three body locations used with four colors (C = 4, L = 3, N = 3) would provide: $(3!/0!3!)4^3 = 64$ unique codes. Recall that $0!=1$.

Get our app!

There's an app for this! NMT's VIE Color Code Generator quickly calculates and lists the unique codes available for a given combination of body locations, tags and colors. You can download a free copy at www.nmt.us. If you are tagging reptiles or amphibians, you may also like to check out Salamarker – another VIE code generator created at Purdue University.





VIE Tag Detection

A powerful feature of Visible Implant Elastomer tags is that red, pink, yellow, blue, orange, and green can be fluoresced for enhanced detection. When fluoresced, a small spot of VIE can be seen at considerable distance and in the dark, and tags obscured by pigmentation are often detectable. However, color perception can vary among samplers. Tags of different colors can generally be distinguished in ambient light in clear tissue, but those same colors may be confused if the marks are poor or are placed under pigmented tissue. Fluorescing the tags greatly enhances tag detection, but red and pink, and green and yellow, look similar when fluoresced.

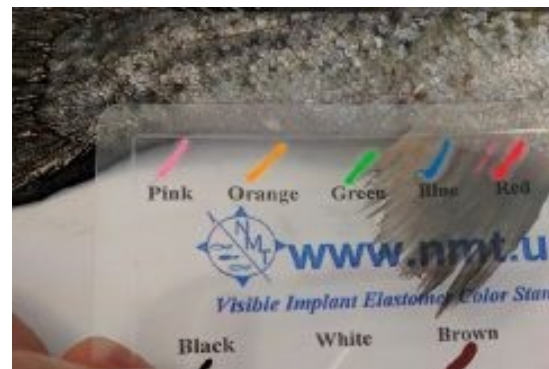
Maximize Tag Identification

- ▶ Choose distinct colors for tagging.
- ▶ Tag in clear tissue whenever possible.
- ▶ Train your samplers – let them practice with the tag colors they will encounter before they start collecting data.
- ▶ Use the VIE Color Standard to correctly identify colors.
- ▶ Use the VI Light to fluoresce poor or obscure tags.

VIE Color Standard

NMT's VIE Color Standard presents the ten VIE colors on a clear card. This allows the sampler to place the color sample directly beside a tag for comparison, either under or over the tagged tissue.

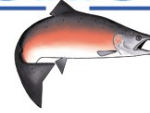
Customized color standards can be made at the time of tagging by dispensing some tags onto clear stiff plastic and then covering them with clear packing tape after the marks have cured. Labeling the colors is helpful.



Using the VIE Color Standard to compare tag colors.

Test the VI Light with the VIE Color Standard to ensure that the tags are adequately fluoresced. Shine the light directly on the area where the tag is thought to be. If you are working in direct sunlight, you will need to fluoresce the tags in the shade – even the shade of your body is probably enough. Very faint tags are best seen when fluoresced in darkness.

CAUTION! Our VI Lights are very bright, but you can hardly see their light. They do not trigger the eye's normal defense mechanisms against bright light. Never stare directly into any light used for fluorescing VIE tags, and keep these lights out of the reach of children.



Fluorescent colors: red, orange, yellow, green, pink, and blue

Non fluorescent colors: purple, brown, white, and black.

Technische Daten / technical details			
Art.no. (AQ)	NMT code		
K0273100	1VIFE000004	VIE Trial Pack	incl. - 1 x 1 ml of elastomer - mixing and injection supplies - written and video instructions
K0273200	1VIFE000002	VIE 6 ml KIT	incl. - 2 x 3 ml of elastomer - 1 x manual elastomer injector - mixing and injection supplies - 1 x VI light - 1 x VIE color standard - 1 x field carrying case - written and video instructions
K0273250	1VIFE000008	VIE 6 ml Refill (with accessories)	incl. - 2 x 3 ml of elastomer - mixing and injection supplies - written and video instructions
K0273260	1VIFE000010	VIE 6 ml Refill (w/o accessories)	incl. - 2 x 3 ml of elastomer
K0273300	1VIFE000007	VIE 24 ml Kit	incl. - 8 x 3 ml of elastomer - 1 x manual elastomer injector - mixing and injection supplies - 1 x VI light - 1 x VIE color standard - 1 x field carrying case - written and video instructions
K0273400	1VIFE000001	VIE 60 ml Kit	incl. - 20 x 3 ml of elastomer - 2 x manual elastomer injector - mixing and injection supplies - 1 x VI light - 1 x VIE color standard - 1 x field carrying case - written and video instructions