

## **Status of DNA Barcoding of Coleoptera, Hymenoptera and Diptera**

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### **Abstract**

DNA barcoding is the taxonomic method that uses a short genetic marker in an organism's DNA to identify it as belonging to a particular species or strain. Although, several loci have been suggested, the most common locus of the animals includes the mitochondrial COI gene. The classical use of morphological trait for species identification has several limitations. They include, for example, the misidentification of a taxon due to the phenotypic plasticity of the trait studied or the existence of cryptic taxa. The DNA barcoding approach might correctly present the best solution for identifying species when their morphology is of limited use. DNA barcoding is promising in the rapid description of biodiversity. Besides adult, DNA barcoding also helps in identification of larvae and pupae. DNA barcoding solves the problem of identification of sibling species, cryptic species and sub species. This paper reviews the status of DNA Barcoding of Coleoptera, hymenoptera and Diptera.

**Keywords:** DNA Barcoding, Mitochondrial COI gene, Coleoptera, Hymenoptera, Diptera

### **Introduction**

Coleoptera is the most diverse order of insects in the world (Footit *et al.*, 2009) and dominates many ecosystems in terms of individual abundance and niches occupied. The common name of **beetles** is used synonymously with Coleoptera, but many have their own common names, such as weevils, beetles, beetles, ladybugs, sanjuaneros, oilers, beetles (in Colombia ), flying deer, fireflies, congorochos (in Venezuela) cáculos (in Puerto Rico ), etc. These organisms exhibit great diversity in terms of species and colonisation of habitats, are easy to sample (Rosenberg *et al.*, 1986) and present high sensitivity to environmental interference. Therefore, the composition of this group reflects the performance of the ecosystem and is fundamental in the analysis of the landscape structure.

Hymenoptera is an important group of insects to man, because they contain pollinators, predators, parasitoids and honey bees which help human beings to improve the economy and agricultural production. They are very essential for natural pest control. So agro ecosystems should have a rich diversity of hymenopteran insects. Hymenoptera is a diverse group of insects in terms of structure, size, numbers, habits and food preference. They are the most evolved and probably most diverse of all the terrestrial organisms (LaSalle *et al.*, 1993).

Flies (Diptera) are among the most omnipresent and widely distributed insects. Their close association with humans has led them to be recognized as unpleasant and disturbing creatures, and certainly, some flies are responsible for millions of illnesses and deaths among human populations. Yet flies are also among the key components in most ecosystems and are advantageous in many ways (Skevington *et al.*, 2002). Insects of this order use only a single pair of wings to fly, the hindwings being reduced to club-like balancing organs known as halteres.

Taxonomic identification of above-mentioned insect orders is male oriented. However, this classification has given rise to many controversies. The morphological identification of these orders is done by the use of male genitalia in combination with more traditional characters of adult. Earlier, some taxonomists used these characters to identify these orders but majority of these names are not valid now. The exact identification up to species, sub species level can be made only with the help of DNA barcoding. DNA barcode is a genetic signature that occurs naturally within the genome of every living species. One of the gene regions commonly used for all animal groups is a 650bp region in the mitochondrial COI gene. The characterization based on morphometric characters is not well suited for phylo-geographical studies because they can be sensitive to environmental selection pressures, need a lot of time and experience, and sometimes are unsuitable for identifying some hybrids. Misidentifications at the level of species and genera have therefore, created serious problems for researchers in the field of ecology, physiology and genetics for a comparison and evolution of their results. Thus, molecular studies have therefore proved to be extremely useful for this purpose.

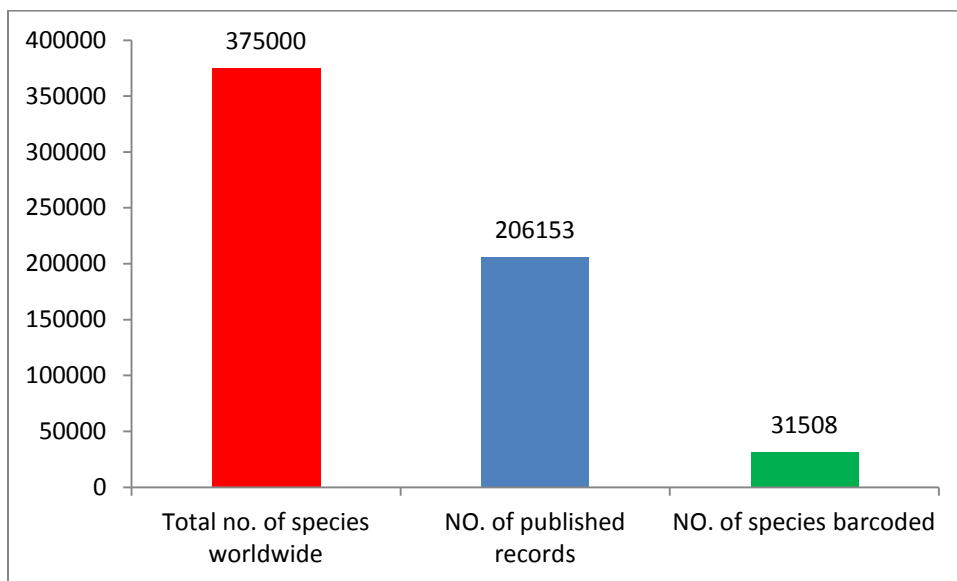
### **DNA barcoding**

The method to identify, label and classify organisms is largely built around morphological characteristics. It was developed by Carl Linnaeus in the 18th century and his taxonomic system is to a large extent still used. Today taxonomists also consider physiology, behavior and population biology in the classification of new species. Since the discovery of DNA and recognition of its role in inheritance, genetic variation plays a major role to distinguish the diversity of life. Morphological identification of species is obviously limited since it does not consider phenotypic plasticity, genetic variation of individuals or morphological complexity (e.g. cryptic taxa or keys only developed for certain gender or life stage) (Hebert *et al.*, 2003). DNA barcoding could fill these gaps and most importantly add new biological diversity to the already known.

DNA barcoding is the taxonomic method that uses a short genetic marker in an organism's DNA to identify it as belonging to a particular species or strain. The latest method of species identification through DNA barcoding of mitochondrial cytochrome oxidase gene I (COI) (Hebert *et al.*, 2003) provides an easy un-erroneous approach for the routine identification of Coccinellid beetles (Fu *et al.*, 2006). Molecular identification and phylogeny using species identification markers such as COI of mitochondrial region is regarded as efficient (Jalali *et al.*, 2015). Mitochondrial markers are used for revealing phylogenetic relationships among related groups, because mtDNA is maternally inherited, it evolves fair rapidly and most of the nucleotide substitutions occur at neutral sites. The main advantage of DNA barcoding is the rapid acquisition of molecular data (Monaghan *et al.*, 2009). DNA barcoding today is done routinely using ordinary PCR and Sanger sequencing. However, a new generation of sequencing techniques, referred as NGS, has been developed in the past decade. The technique combines DNA capturing, PCR and sequence reaction, and hence could be suitable for DNA barcoding.

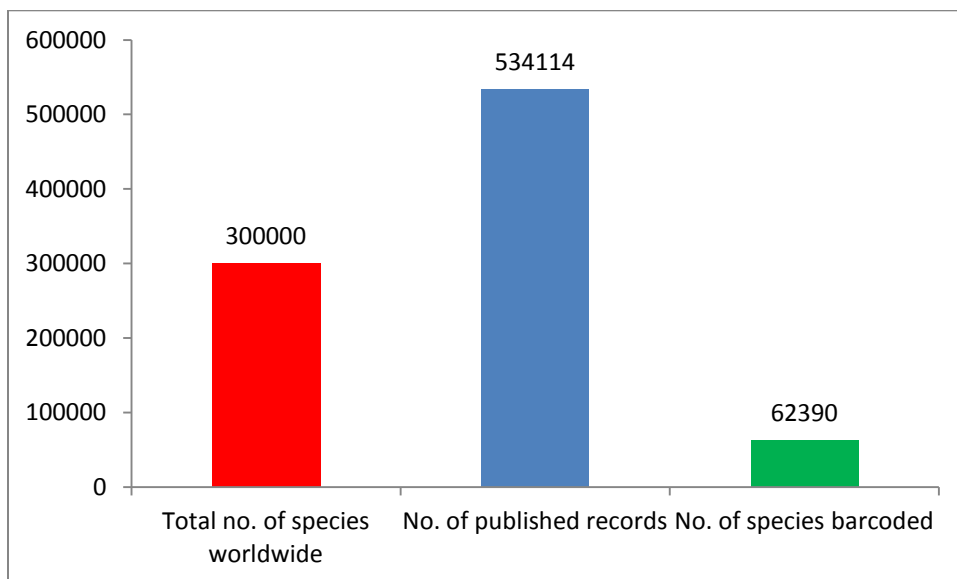
### **DNA barcoding of three economically important insect orders**

**1. Coleoptera:** Coleoptera is an order of insects with about 375,000 species described; it has many species such as vascular plants or fungi and 66 times more species than mammals .it contains more species than any other order throughout the Animal kingdom , followed by Lepidoptera , Hymenoptera ( bees , wasps and ants ) and Diptera ( flies , mosquitoes ). Coleoptera is the largest order of insects, with about 400 thousand species worldwide, representing 30% of animals and about 40% of all insects (Lawrence and Britton, 1991). So far only 31508 species have been barcoded (**Figure 1**).



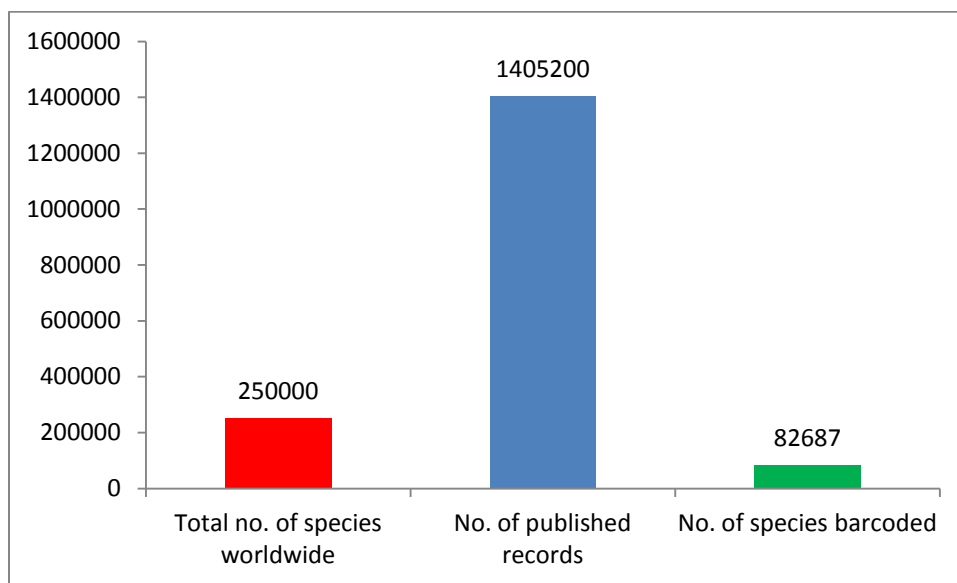
**Figure 1: Showing details of DNA barcoding of Coleoptera worldwide**

**2. Hymenoptera:** Hymenoptera is one of the few mega diverse insect orders. Approximately 300,000 species are estimated to be present worldwide and nearly 115,000 species of hymenoptera have been described so far (Grissell, 1999). Parasitic hymenoptera constitutes nearly 25% of all arthropods in both temperate and tropical ecosystems (Stork *et al.*, 1988). The order hymenoptera has nearly about 300,000 described spp. out of which only 62390 spp. have been successfully barcoded (**Figure 2**).



**Figure 2: Showing details of DNA barcoding of Hymenoptera worldwide**

**3. Diptera:** Diptera is a large order containing an estimated 25,000 species including horse-flies, crane flies, hoverflies and others, although only about species have been described. The name Diptera (commonly known as true flies) was originally assigned by Aristotle and adopted by Linnaeus in 1744. Mosquitoes, flies (black, fruit, blow, house etc.), midges (biting, non-biting), gnats (fungus, root), keds, bots etc., are the members of the order Diptera. They are very minute to large insects (0.5-75 mm long), variable in colour and form, soft bodied and the adult flying forms with only one pair of mesothoracic wing and with a pair of modified hind wing i.e. haltere (Banerjee *et al.*, 2006). Out of reported species only 82687 have been barcoded (**Figure 3**).



**Figure 3: Showing details of DNA barcoding of Diptera worldwide**

### Conclusions

If DNA barcoding proceeds on a large scale, it will generate important by-products for the scientific community. All DNA extracts produced during the barcode analysis of vouchered specimens will be stored, allowing future efforts to examine patterns of sequence diversity in other gene regions, and the collection programs instigated by DNA barcoding will expand the specimens available for morphological analysis. The barcode initiative will also create a Web-based system delivering not just automated identifications, but also providing a portal to biological information for all species included in the registry. Although DNA barcoding will not create the “encyclopedia of life,” it will generate its index and table of contents. We view these signs of growing synergy among the various sectors of the biodiversity community as extremely hopeful. If developed to their full potential, history may view the DNA barcoding enterprise as one that not only enhanced access to taxonomic information, but also strengthened alliances among all those with interests in the documentation, understanding, and preservation of biodiversity—an exciting prospect indeed.

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