

Assessment Of DNA Isolation From Oven Drying Preserved *Eudocima phalonia* And *Hippotion rosetta* Body Parts.

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ABSTRACT

Great diversity and huge variety of insect is observed in kingdom Animalia. The order Lepidoptera has strong genetic background, which has a wide scope for further studies. Thus, apart from basic molecular studies, DNA studies are also important for study in the diversity of insect. The present study confirms isolation of variable amount of DNA from different body parts of moths. The species of moths are collected from Saptshrungigud, Vani-Nashik (MH) at night time and preserved it by oven dried method at around 57⁰c temperature overnight. Here, the total genomic DNA was isolated from individual adult moth's leg, head, thorax and abdomen by using phenol-chloroform extraction method and colorimetric quantification of isolated DNA was done by diphenylamine (DPA) method. The result; propose that leg can serve as preferred tissue for more DNA isolation from *Eudocima phalonia* and *Hippotion rosetta* body parts.

Keywords: Lepidoptera, DNA studies, *Eudocima phalonia*, *Hippotion rosetta*.

INTRODUCTION

The insect diversity is enormous and is of wide range. Lepidopterons account for a great number amongst all insect. Lepidoptera is large order of insect that include Moths and Butterflies. It shows many variations in basic body structure, which have evolved to gain advantages in life style and distribution. They are morphologically distinguished from other order, principally by the presence of scales on the external body parts and appendages.

Moth's shows great diversity of variety in wing pattern observed which has genetic background. As a result make molecular studies an important tool for studying diversity. In order to study the genetic correlation with diversity among these species, it is proposed to study the DNA profile for which DNA isolation is the first and most crucial step. Therefore, a genetic study on moths has wide scope.

Each and every cell contains DNA and thus theoretically any cell or tissue can be used for DNA isolation. On the basis of above mention reasons, we have assessed body parts for serving as ideal tissue for DNA isolation from moths. This assessment was done on *Eudocima phalonia* and *Hippotion rosetta* species of moths belonging to family Noctuidae and Sphingidae respectively.

MATERIALS AND METHODS

Collection and Preservation: Moths were collected from Saptshrungigud Vani-Nashik (MH) at night time with the help of insect collection nets. All collected moths are killed with chloroform in killing jar. The moths are then spread evenly on insect spreading board followed by oven dried at 57⁰c temp overnight.

*Eudocima phalonia**Hippotion rosetta*

DNA Isolation: the DNA is isolated from preserved moth's body parts like head, thorax, abdomen and leg by using Chloroform: iso-amyl-alcohol (24:1) and phenol. Total genomic DNA was extracted from individual adult moths body parts by a modification of technique of Harrison et.al. described by Sperling et.al.

DNA Quantification: Colorimetric quantification of isolated DNA was done by using diphenylamine method. Diphenylamine reagent reacts with DNA to produce a deep-blue coloured compound which was read at 620 nm. The standard values were plotted and concentrations of DNA isolated from body parts.

RESULT

To assess amount of DNA isolated from different body parts was significantly different. In these two species under study, it was found that the amount of isolated DNA in *Eudocima phalonia*'s Leg (200 µg), Head (100 µg), Thorax (72.5 µg) And Abdomen (87.5 µg) and in *Hippotion rosetta*'s Leg (170 µg), Head (115 µg), Thorax (80 µg) And Abdomen (60 µg) by colorimetric quantification By diphenylamine method from 50 mg of tissue. It can be determined all the observations of the isolated DNA the maximum DNA was isolated from leg.

Body Parts	<i>Eudocima phalonia</i>	<i>Hippotion rosetta</i> .
Leg	200	170
Head	100	115
Thorax	72.5	80
Abdomen	87.5	60

Table : Mean values of amount of DNA isolated (µg) from 50 mg of tissue

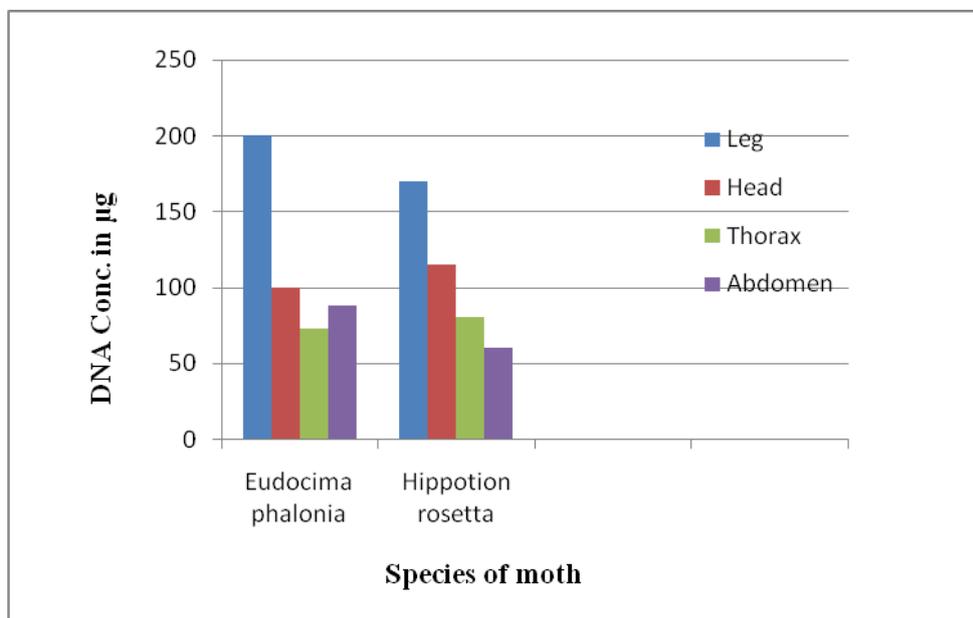


Fig. : Comparison of DNA isolated from various body parts of different species.

The mean amount of DNA (μg) isolated from leg was 200 ± 8.16 for *Eudocima phalonia* and 170 ± 8.16 for *Hippotion rosetta*. The data obtain from comparative analysis of different body parts for assessing the tissue in given table. This shows that maximum DNA was isolated from leg as compare to head, thorax and abdomen in both species under study.

DISCUSSION

The amount of DNA isolated from different body parts is variable. This is confirmed by statistical tool ANOVA applied on the data obtain in the present study. The mean amount of DNA is isolated as in *Eudocima phalonia* $72.5 \mu\text{g}$ mean amount of DNA was isolated from thorax and $200 \mu\text{g}$ mean amount of DNA was isolated from leg Which was 2.76 times greater as compare to thorax and *Hippotion rosetta* $60 \mu\text{g}$ mean amount of DNA was isolated from abdomen and $170 \mu\text{g}$ mean amount of DNA was isolated from leg Which was 2.8 times greater as compare to abdomen.

Lepidopteron have characteristics features of presence of scales on the body surfaces. Leg is the only body part considered in the present study which do not has scales over its surface. The 50 mg of tissue taken for DNA isolation is composed of two parts; i.e. cells and scales. More the cells more would be the amount of DNA isolated. Thus, if minimum scales are present in the tissue taken for DNA isolation more would be the amount of DNA isolated. Therefore, maximum DNA isolated from leg as compared to other body parts.

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