Chapter VI
Summary &
Conclusion
Though a lot of information is available on the molecular aetiology of male factor infertility from different parts of India and abroad, there is a complete lack of information on the role of mtDNA mutations in male factor infertility in Northeast Indian population. Even sufficient data is lacking regarding the prevalence of Y Chromosome microdeletions in infertile males of this region of India. In view of direct relevance of this information to the treatment and management of male infertility with different grades of severity of condition the present study is undertaken on infertile males of Northeast India.

In this study a total of 330 infertile males belonging to nine different categories of male infertility are included in the test group. In addition 50 childless, normozoospermic males are randomly selected as control individuals. About 86 individuals from test group are randomly selected for mtDNA mutation analysis. For this purpose 20 childless, normozoospermic individuals are included in the control group. The salient features of the study are summarized below:

1. The prevalence of Y Chromosome microdeletion in different categories of infertile males belonging to test group is found to be higher than the control group.

2. The frequency of Y Chromosome microdeletion among infertile males is found to be 18.18% compared to 2% in control group.

3. Among infertile males, the frequency of Y Chromosome microdeletion is found to be 31.67% in AZFa region (at least one marker is deleted), 33.33% in AZFd region (at least one marker is deleted) and 35% in AZFa + AZFd region (at least one marker from each region is deleted).

4. The frequency of Y Chromosome microdeletion in semen sample analysis is found to be higher than the blood sample analysis.

5. The prevalence of Y Chromosome microdeletion in azoospermic cases is found to be higher than the oligozoospermic cases (Table-IV.5).

6. In comparison to the control as well as oligozoospermic group (p<0.001), the azoospermic males have significantly lower serum testosterone level.
7. The serum estradiol level in azoospermic males is found to be significantly different than the control group (p<0.001), but insignificantly different from oligozoospermic males (p>0.001).

8. The serum prolactin level in azoospermic group is observed to be significantly higher than the control group (p<0.001), but insignificantly different from oligozoospermic males (p>0.001).

9. The semen pH value in all the groups is observed to be between 8-9.

10. The serum testosterone level of control group is found to be within the normal range but all the different test groups consisting of 330 individuals have values below the normal range.

11. All the infertile groups showed lower serum testosterone level but significantly higher serum estradiol level than the control group (p<0.001).

12. The serum prolactin level of all the test groups is found to be apparently higher than the control group (Table-IV.3).

13. Among the men with microdeletion of Y Chromosome, 66.67% showed microdeletion of AZFa region and 68.33% have shown microdeletion of AZFd region either individually or in association with other markers.

14. The frequency of microdeletion of USP9Y gene (AZFa region) is found to be highest in asthenozoospermic men (10%) and that of DBY gene (AZFa region) is observed to be in oligoteratozoospermic group (16.67%).

15. Of the three STS markers of AZFd region included in the study, highest frequency of microdeletion of sY145 is observed in oligoasthenoteratozoospermic group (9.09%), highest frequency of microdeletion of sY153 is observed in oligozoospermic group (13.41%) and highest frequency of microdeletion of sY152 is observed in cryptozoospermic and oligoasthenozoospermic group (11.11%).

16. In the present study, six candidate genes of mtDNA are included for mutation analysis and among them highest number of point mutations (involving base substitution) i.e., single nucleotide polymorphisms (SNPs) are found in ATPase6 gene (21 SNPs), followed by ND2 gene (12 SNPs) and ATPase8 gene (9 SNPs).

17. In total 50 different SNPs are observed in these six genes of which 24 are detected for first time in infertile male cases.
18. In the study population, haplogroup Y is found to be predominant among the infertile male cases.

**Limitations and Future Scope:**

This is the first study on the role of mtDNA mutations in male infertility involving infertile cases of Northeast India. Although this study is able to throw light on some important aspects in this regard, it has some limitations. Although best efforts are made to collect as many cases as possible under each category of male infertility, but our efforts are hindered due to unavailability of sufficient number of cases in the infertility clinics from where the samples are collected. Therefore, many infertile patient groups of the present study have very low number of cases. As the study is based on the cases visiting the infertility clinic of Guwahati, a large population of infertile males from other parts of Northeast India are left out of the study. The study has the aim of determining the prevalence of microdeletion of only AZFa and AZFd regions of human Y Chromosome and other two important portions of AZF region are not included which has left a scope to determine the prevalence of co-occurrence of microdeletions in all these four regions. Also due to limitation of funding and other operational problems only 6 candidate genes from the mtDNA are included for the mutation analysis.

However, the outcome and limitations of the present study has opened several doors for further research in this area.

1. A detailed study including all the four portions of AZF region can better elucidate the prevalence of Y Chromosome microdeletion in the study population.

2. It also lefts us with the scope of determining the role of environmental and lifestyle related factors in male factor infertility.

3. Determination of the role of mutations in all the mtDNA genes encoding the enzyme complexes of electron transport chain can also help in better understanding of the etiology of male factor infertility in Northeast India.