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Scientific method of preparing homoeopathic nosodes

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ABSTRACT

Introduction: Nosodes are homoeopathic preparations sourced from biological materials such as diseased tissues, organisms, cultures (bacteria, fungi, and viruses), or parasites, or from decomposed products from humans or animals. More than forty-five major nosodes have been in use since 1830 but no clear guidelines regarding their preparation are available.

Objective: To standardize the method of preparation of nosodes using modern technology and lay down clear guidelines for the same.

Materials and Methods: Biological material identification such as culture of organism, separation of required pure fraction, quantification, standardization, dilutions, potentisation method, and safe use of nosodes were documented in a systematic way.

Result: HIV, HCV and Mycobacterium tuberculosis nosodes were prepared using the modified method described in this article.

Conclusion: The improved method of nosode preparation can pave way for laying clear guidelines for preparation of nosodes in homoeopathic pharmacopeias.

Keywords: Ethics, Human immunodeficiency virus, Hepatitis C, Method, Mycobacterium tuberculosis, Nosode, Organism, Potentization, Standardisation

INTRODUCTION

Nosodes are broad-spectrum, widely used homoeopathic preparations sourced from biological materials such as cultures or clinical samples of microorganisms (e.g. bacteria, fungi, and viruses) or from parasites, diseased tissues (cancerous tissues), or decomposition products from humans or animals. About forty-five[1] such nosodes have been used by homoeopathic practitioners since 1830.

Discovery of itch mite in 1687 marked scabies as the first disease of humans with a known cause.[2] It is interesting to note that *Psorinum* (a nosode from the content in scabies vesicle) was the first nosode sourced from infected material, and was prepared and proved by Hering in 1830.[3] Most of the other nosodes were made between 1875 and 1925. Understandably, they were not standardised in terms of source, quality, quantity, purity, count, morphology, characterization, and safety. The homoeopathic pharmacopoeia was evolved from 1825 to 1950.[4] The Homoeopathic Pharmacopoeia of the United States (1901) prescribed guidelines for making nosodes. All the major nosodes (*Psorinum, Medorrhinum, Syphilinum, Tuberculinum, Bacillinum, Carcinosin, Pyrogen, Lyssin, Variolinum*) were prepared before 1901. The symptoms for most of the nosodes as we find in different source books like HC Allen's Materia Medica of Nosodes were derived from clinical cases as there were scanty

drug-provings conducted. (Table 1: Overview of the history of nosodes).

Microbiology and standardization techniques in the study of bacteria, virus, fungi, and parasites evolved significantly in the past 50 years, after which the nosodes were not re-made. No major new nosodes (except Oscillococcinum and Leptospirosis) were introduced from newer organisms such as Human Immunodeficiency virus (HIV), Hepatitis C, polio, herpes, leprosy, and dengue.

OBJECTIVES

To standardize the method of preparation of nosodes using modern technology and lay down clear guidelines for the same.

RATIONALE

Attention must be drawn to the fact that it is almost impossible to remake any of the existing homoeopathic nosodes, as they were prepared by Hering[3], and others from obscure sources over hundred years, without standardisation of the source materials, For example, *Psorinum* was prepared *around 1828* from sero-purulent matter contained in the scabies vesicle.[5] It must be noted that the purulent matter must have contained unknown organisms, proteins, and debris, besides suspected organisms (*Sarcoptes scabiei*) responsible for scabies. Similarly, before 1890, *Medorrhinum* was prepared from the urethral discharge of the patient having Gonorrhea,[6] which must have, similarly, contained more than pure *Neisseria gonorrhoeae* organism, as microbial culture facilities were primitive at that time. It can be suspected that the so-called gonorrheal discharge may have had mixed infection of other organisms (including other venereal diseases), proteins, and debris. One more example of *Tuberculinum* sourced from the mucous of a patient suspected to have tuberculosis[7] by Swan in 1879, or from the lung tissues of a patient suspected to have suffered from tuberculosis, or from Tubercular glands from slaughtered cattle[8] led us to understand that the source material used in *Tuberculinum* must have contained substances much more than the pure *Mycobacterium tuberculosis* organism, which were found by Robert Koch in 1882.[9] *Syphilinum* was prepared by Swan around 1880 from *Syphilis (*earlier called virus*)*[10] while *Treponema pallidum* was discovered in 1905 by Schaudinn and Hoffmannin.[11] These examples of four major homoeopathic nosodes, whose symptoms have been described in the Materia Medica, convey that there is no way to re-produce them.

We can, of course, re-make the nosodes, which could be now sourced from standardised source material. However, in that case, the question will arise: Can they be prescribed based on the symptoms derived from proven drugs that were conducted from incomparable source material over 100 years ago?

Not many nosodes were systemically prepared and introduced to the profession in past 100 years. *Oscillococcinum* made from the heart and liver of wild duck,[12] which is a patented homoeopathic preparation manufactured by Boiron Laboratories, was introduced to the profession, but there is no literature available about its Homoeopathic Pathogenetic Trial (HPT). Similarly, no data are available about the standardised source, HPT, or clinical trials for some nosodes such as leptospirosis nosode or Malaria Co.[13]

After 1990, Chikungunya became rampant and attacked the population in an epidemic form in India and other countries. Two nosodes, one from the whole mosquitoes and another from the blood of an affected patient of Chikungunya, were prepared.[14] Sources such as whole mosquito or blood of infected patient cannot be considered standardised. Isolated virus strain of Chikungunya is now available, and fresh nosode can be prepared using viral culture. A trial conducted in 1996 by Central Council of Research in Homoeopathy (CCRH, News Vol. No. 23, Scientific Activities of Council, Epidemic Dengue), the Dengue Fever nosode (source: Serum of a person suffering from dengue fever) in 30C potency was administered to people in Delhi during an epidemic of Dengue haemorrhagic fever. In all, 39,200 people were given the nosode; 23,520 were successfully followed up, and only five people had developed symptoms. There was an infection rate of 0.125%, compared to what was normally expected, about 50%.[15,16] In 2007, the Finlay Institute in Cuba prepared a Leptospira nosode 200 CH using four circulating strains and used as homoeopathic prophylactic.[17]

The nosodes available in the homoeopathic market have been prepared possibly from old back-potencies, sources of which have not been described and cannot be tracked correctly.

Table 1: Illustrations of the history of few major nosodes

Nosode	Source, substance	Prepared by	Year of preparation	Year when organism found	Systematic drug proving conducted	Reference
Tuberculinum	Sputum of suspected tuberculosis (Lung tissues)	Swan	1879	*Mycobacterium tuberculosis*, 1884, by Robert Koch	Not a single mental symptom is from drug proving[7]	Hering guiding symptoms[7]
	Pus (with bacilli) from tuberculosis abscess			*Mycobacterium tuberculosis*, 1884	No	Allen HC, Keynotes[18]
				Mycobacterium tuberculosis, 1884	No	Boericke W. Pocket Manual of Homoeopathic Materia Medica[19]
Tuberculinum-Bovinum	Tubercular glands from slaughtered cattle	Kent used one made by Boericke and Tafel[20]	Before 1910	*Mycobacterium tuberculosis*, 1884	No	Kent's Materia Medica[8]
Bacillinum	A maceration of a typical tuberculous lung	Burnett	1885-90	*Mycobacterium tuberculosis*, 1884	No	Boericke W. Pocket Manual of Homoeopathic Materia Medica[20] Sarkar BK.[21]
Syphilinum	'Syphilitic virus.' No source specified	Swan	1880	*Treponema pallidum*, 1905 by Schaudinn and Hoffmannin,[10]	No	Hering C. Guiding symptoms[10]
Medorrhinum	The Gonorrhoeal virus	Swan	1880-85	*Neisseria gonorrhoea*, 1879, by Albert Neisser	No	Boericke W. Pocket manual of Homoeopathic Materia Medica[22]
Psorinum	1. Sero-purulent matter contained in the scabies vesicles used for proving (No information on the person making it and the year) 2. Epidermoid efflorescence of pityriasis used by Gross 3. The salt form the product of Psora, used by Hering	G.W. Gross (1794-1847), Hering (around 1830) (Exact source of the current preparation not known. Unclear as to what is meant by the salt of the product of Psora)	1830	Itch mite was found in 1687. Diacinto Cestoni discovered Sarcoptes before 1718	Yes (There is no clarity about the source used in the drug-proving)	Hering C. Guiding symptoms[3,20] No mention of Psorinum in Materia Medica Pura (1825) and Chronic Diseases (1828)
Pyrogenium	Septic pus	Swan (Unclear whether it contains secretions of Gram positive, or negative, or both)	1887	Gram positive and negative bacteria were identified by Hans Christian Gram in 1884	No	Boericke W. Pocket Manual of Homoeopathic Materia Medica[23]

MATERIALS AND METHODS

With the advancement of microbiology, immunology, and cellular biology, it is now essential to standardise the therapeutic source material for preparing nosodes and a revised potency preparation parameters needs to be rewritten, which can help in the following ways.

After having identified the need of modern scientific era, the proposed methodological procedure of nosode preparation is as under:

- The standardized method of preparation will be reproducible if the guidelines laid down are followed by the pharmacies
- In case of clinical sample, consent from the donor has to be documented.

Steps to Nosode Preparation

The steps have been described herewith giving example of one of the nosodes prepared using this method.

Step I: Identification and procurement of source material

It is necessary to identify and document the authentic source material. Standard tests must be specified confirming the exact organisms. In case of HIV, HIV duo serum screening test (fourth generation test), and in case of Hepatitis C, Hepatitis C antibody test may be done to confirm the specific organisms. Research purpose and interest of commercial application must be clearly stated in the consent. In case of organism (may include bacteria, virus, protozoa, parasites, or fungi) or other biological material sourced from academic or commercial source, it must be documented accordingly.

- Source material/Strain: To use the latest virulent or standard strains of organisms (culture, whenever possible), resistant strains, combinations of various strains or using fresh clinical samples of diseased material, biological material when culture or isolation is not possible
- Organism count: The count of the organism in the source material or in the mother source must be specified, achievable, and significant. The recommended organism count for making the nosodes suggested in HPI (Homoeopathy Pharmacopeia of India) is 20 billion.[24] However, it may not be possible with many organisms (especially virus, fungi, parasites, and some bacteria) to achieve such a high count. For example, *M. leprae* organisms cannot be cultured *in vitro* to achieve large count. Based on the nature of organisms, the count may be specified in individual monographs
- Ethical consideration: In case the blood samples are drawn from volunteers, informed consent form must be served to him/her declaring the proposed use of the sample. Ethical guidelines for biosafety,[33] viral safety of human blood plasma product, safety issues in preparation of homoeopathic medicines and clinical research are now in place.[34] World Health Organization formulated guidelines for biosafety in the form of Laboratory Biosafety Manual and for clinical research in the form of Declaration of Helsinki in 1964. The Indian Council of Medical Research has specified the 'Ethical Guidelines for Biomedical Research on Human Subjects.[35]

Step II: Nature of material

Depending on the nature of material, whether organisms are capable of producing endotoxins, exotoxins, made from purified toxins or made from microbes, viruses, or clinical material from diseased subjects, these preparations are divided in HPI into four groups[24] N-I, -II, -III, and -IV.

N-I–Preparations made from lysate of micro-organism capable of producing bacterial endo-toxins; *e.g. Salmonella* Typhimurium, *Escherichia Coli*, and *Staphylococcus*.

N-II – Products made from micro-organisms capable of producing exotoxin, *e.g. Corynebacterium diphtheriae.*

N-III – Preparation made from purified toxins.

N-IV – Preparation made from micro-organisms/viruses/clinical materials from human convalescents or diseased subjects, *e.g.* Variolinum, Influenzinum, Psorinum, Syphilinum, and Morbillinum. New nosodes sourced from HIV, Hepatitis C, and *Mycobacterium tuberculosis* fall under this group.

Step III: Removal of common co-infection/contamination

Purity of the source material has to be established. All possible contaminants must be removed. In case of the source material being blood samples, they must be tested to rule out other known possible co-infections. In cases of Hepatitis C and HIV nosodes, possible co-infections such as gonorrhoea, syphilis, herpes, and tuberculosis were ruled out. In case of pure cultures, this step may be omitted.

Step IV: Removal/Separation of other components

In case of any nosode sourced from serum, serum expression, centrifugation, and/or filtration can be used to procure the organism from the source material. We had subjected the blood samples collected from HIV and Hepatitis C positive donors to serum expression to separate blood cells and

suspended particles from the whole blood. The samples were subjected to centrifugation to obtain clear serum and filtration (Seitz filter) to get rid of cell debris, unidentified bacteria, and large protein particles. If the source material is obtained as scraping of parasite-infected animal or human tissues, for example, in case of *Sarcoptes scabiei* the pure parasite is to be isolated. In such sample, the keratin component of skin is to be removed by boiling the scrapings with potassium hydroxide (KOH) in water medium.

Step V: Characterization of source material

The microorganisms need to be characterised in terms of genotyping and strains, as per the latest available technology. In case of bacteria-strain characterization and for virus, typing requires to be done. We had included genotype I and III in preparation of Hepatitis C nosode, and type I and II for HIV nosode. In case of leptospira, serogroups, serovars, and genomospecies, and for fungi, types such as mould, yeast, or mushroom must be specified. Similarly, identification of the toxins, endotoxin, or exotoxin can be documented. For example, diphtheria toxin is a protein exotoxin produced by *Corynebacterium diphtheriae*, Pertussis Toxin (PT) is a protein-based AB5-type exotoxin, and Cholera Toxin (sometimes abbreviated to CTX, Ctx, or CT) is a protein complex secreted by the bacterium *Vibrio*.

Step VI: Safety

Biosafety: Stringent biosafety compliant environment is recommended with minimum handling, using sealed containers and disposable auto-tip pipettes.

The safety of nosode in various potencies must be established as per the sterility testing mentioned in Indian Pharmacopoeia or European Pharmacopoeia[29] for aerobic and anaerobic organisms.[31] Sterility testing results indicating the presence and growth of source organisms in 1C, 2C, 6C, etc., if any, must be documented, establishing the absence in a specific potency and onwards. Any potency below 6X should not be dispensed and the potencies above 6X must be sterile as per HPI.[24] Heat inactivation of pathogens[32] must be documented if carried out. However, the inactivation is not suggested by HPI.[24] The author has opted for live organisms (without heat inactivation) as source while preparing HIV, Hepatitis C, and *Mycobacterium tuberculosis* nosodes to avoid loss of virulence by the process of inactivation.

Step VII: Mother preparation

At this stage, specified quantity of pure culture of one strain or more (polyvalent) nosode or more than one type of organisms can be mixed in vehicle to obtain original stock nosode. In case where nosode is to be prepared using pure culture where there is documented evidence for its purity, one can skip steps III, IV, and V.

- Vehicle: Water for injection that is free from organisms, pyrogens, and NaCl is recommended as preferred vehicle, at least up to 6C potency, as a study by the author has shown presence of source (*Mycobacterium tuberculosis*) organisms up to 5C potency.

In Homoeopathic Pharmacopeia of India[28] and French Homoeopathic Pharmacopeia,[29] alcohol has been advocated as a medium for preparing nosodes. In a small experiment suggested by our team of experts, it was observed that alcohol when mixed with serum (and heated), leads to denaturation of proteins, distortion from the original form, and probably loss of antigenicity. A study has shown that viral antigenicity is affected by the use of ethanol.[30]

Step VIII: Quantification

It should be considered mandatory to specify the strength of stock nosode, whenever technology is available. Organism count is to be done by the digital counters, turbidity match method, or as specified in individual monograph. In case of preparations sourced from virus standardization with respect to viral copies by PCR (Polymerase Chain Reaction) method to confirm that serum had retained specific count of viral copies, and hence efficacy or antigenicity to the product is retained. It is also recommended to document the organism count in 1c potency. Author has documented the 4,153 viral copies by PCR method in HIV type 1 (excluding HIV type 2 viral copies as the method for its quantification is currently (2010) unavailable), 3,39,642 viral copies collectively of Hepatitis C type I and III and in Mycobacterium nosode, 20 billion, by turbidity match method.

Step IX: Potentization: Machine and method

Succussion or trituration: The method of potentization is decided depending on the nature of source material.

- In case where the size of biological material is macron or visible to naked eye, trituration process can be followed

- Trituration of bacteria with Saccharum lactis (the diluent) (particle size of the vehicle is far larger than the size of microbial cell) may not result in further size reduction
- Trituration method has been recommended if disease tissues (such as cancerous organ tissues) were used as source material instead of culture of organisms
- Succussion is recommended over trituration for the microorganisms that are found in micron size, which are generally uniformly spread in the solution, which should be applicable to nosodes sourced from virus, bacteria, and fungi
- European regulations[25,26] recommend the use of succussion for nosodes
- Oscillococcinum (Boiron, France) was prepared from heart and liver extract of duck by succussion.[12,27]

During trituration, for example, *Mycobacterium tuberculosis*, the person who will execute the process and environment will get exposed to such huge count. The trituration method entails transferring of vehicle powder from one mortar to another by scrapping, which leads to the very high risk of exposure. Succussion allows transfer of liquid from a sealed bottle to another bottle using the syringe while making the serial dilutions in controlled environment, which is a safe method.

There is a scope for improvement in the conventional method of potentization with regard to the machine parameters:

Force Parameters

One of the most important genres that homoeopathy is distinguished from all other systems of medicine is through its method of potentization. The most important element in the process of potentization is the *power* exerted by administering strokes to the medicine (bottle) at every step. Logically, the strength of the stroke will influence the efficacy of the medicine. However, we have no standardised parameters defined in the conventional method of potentization, for the strength of the strokes to be administered. With the help of experts, the author has standardised the force parameters for electro-mechanical potentiser machine with following considerations:

i. Machine: Standardization
   Weight of arm: 7.5 Kg
   Arm length: 55 cm
   Angle: 90 degree
ii. Force = Weight x Gravitational acceleration
   = 7.5 kg × 9.8 = 73.5 Newton
   Impact: Torque (Moment of Force)
   Torque = (tau) = τ = r x F = 0.55 × 73.5 = 40.43 Nm (Newton meter) × 10 strokes = 404.3 Nm
iii. Strokes: Number of strokes applied at each step of potentization was ten, which was counted with a digital device attached to the machine, avoiding human errors.

It may be noted that even if there is a change in the above parameters while preparing any new preparation or potency, it is scientifically indicated to have the documentation in place. Uniformity of impact is possible with the mechanical device, which can never be achieved in hand-made potencies.

Preparation of 1C Potency

Required amount (1:9 in decimal potency and 1:99 in centesimal potency) of original stock nosode is to be diluted with vehicle to make 1C potency. Instead of using ancient drop method, use of micropipette is recommended to measure the exact quantity of the stock. Author has taken 0.03 ml of stock and 2.97 ml of vehicle to achieve 3 ml volume of centesimal potency. Choice of vehicle and the method for potentization succussions or trituration may depend on the nature of the source material, as discussed earlier in this article.

1C, 30C, 50C, and More

Subsequent potencies to be prepared using the specified potentiser, with the proportion of 1:9 or 1:99 for decimal and centesimal potencies, respectively, as per the method described in Homoeopathic Pharmacopeia of India.[24]

Step X: Safety check for human use

To safeguard the human subjects' participation, it is required to establish the safety of the nosode. As per the HPI, test for sterility as mentioned for aerobic and anaerobic organisms in IP 1964 should be made before issue of any nosode, 6X or below, for therapeutic use or for manufacturing of higher attenuations. It is advisable to use the sterility testing method (*e.g. Mycobacterium Tuberculosis* nosode) as per pharmaceutical industry standards in IP 2010. Additional precautionary measures such as DNA material testing, pro-viral DNA test, ultra-electron microscopy, viral copies Reverse Transcription Polymerase Chain Reaction (RT-PCR) method with positive and negative control would be worthwhile. HIV and Hepatitis C nosodes were subjected to all the above listed testing for two different batches. 30C potency was spiked with serum (containing detectable viral copies) and was used as positive control.

Step XI: Lyophilization

It is suggested to get lyophilisation of the original stock to allow remaking of the nosodes in future, without any need to repeat initial steps. In future, a centralised depository system could be made where standardised raw materials could be preserved for use.

RESULTS

Author has prepared three nosodes (HIV, Hepatitis C, and *Mycobacterium tuberculosis)* considering the above parameters and the steps defined herein.[36] For making the polyvalent nosode of *Mycobacterium tuberculosis*, the author used *Mycobacterium tuberculosis*, *Mycobacterium bovis (BCG vaccine strain)*, *Mycobacterium avium,* and *multidrug resistant Mycobacterium tuberculosis strains.* The projects were approved by the ethics committee constituted and functioning as per the ICMR (Indian Council of Medical Research) guidelines. Safe use of the nosodes was documented with respect to nonexistence of viral copies, DNA material using RT-PCR, and ultra-electron microscopy.

The HPTs and clinical trials for HIV and Hepatitis C nosodes as per the 'Guidance on Good Clinical Practice' (GCP),[37] CCRH (Government of India),[38] and ECH (European Committee for Homoeopathy) guidelines were carried out.[39] The results of the trials have shown some findings for application of these nosodes in certain therapeutic areas. Polyvalent nosode using different latest strains of *Mycobacterium tuberculosis* has been prepared, and this nosode is presently (October 2013) under evaluation process for safety and efficacy studies.

DISCUSSION

Nosodes can be considered as vaccine-like preparation due to this nature of making, where the vaccines are attenuated while the homoeopathic preparations are potentised. However, nosodes are not to be used as substitutes to the vaccines but as per homoeopathic and isopathic principles for the treatment of acute and chronic diseases., The published position paper in 2006 by European Coalition on Homoeopathic and Anthroposophic Medicinal Products[29] reveals the significance of nosodes in Homoeopathy. A 2005 survey conducted by ECH among their members, predominantly homoeopathic doctors, veterinaries, and dentists, revealed that 95% considered nosodes important in their clinical practice.[29] A study by the VKHD (Verband Klassischer Homõopathen Deutschlands) in 2005 of 200 practitioners reported an average of 33% of patient cases where a nosode turned out to be of vital importance for a successful homoeopathic treatment of chronic ailments.

A study with Leptospirosis nosode in in Cuba demonstrated positive effects.[40] The homoeopathic prophylactic approach was associated with a large reduction of disease incidence and control of the epidemic.[17] HIV and Hepatitis C nosodes were developed by the author, which underwent HPTs[36] and clinical trials. In the clinical trials (work not yet published), the HIV nosode has shown some increase in CD4 count (in case of HIV). The Hepatitis C clinical trial has demonstrated decrease in viral load (in Hepatitis C) in an unpublished study, suggesting further research. More nosodes need to be prepared and worked upon, calling for overhauling of the old method and upgrading it with scientific technique.

The improved method of nosode preparation used by the author has demonstrated consideration of aspects such as source material, characterization, vehicle, potentization, force parameter, ethical views, and the safety measures. Safety of the oral use of the nosodes (HIV and Hepatitis C[36]) was established through pathogenetic trials in healthy volunteers and clinical trials.

Having identified that previously prepared (over 100 years ago) nosodes are in use today, without any revision, sourced only from the back-up potencies, there is a need of a serious review. Even if new nosodes are re-prepared, say new Medorrhinum from pure culture of *Neisseria gonorrhoeae* organisms or new Syphilinum from *Treponema pallidum*, they will require fresh HPT (drug proving), as their clinical indications derived from new drug-proving are likely to be different from those listed in the old Materia Medica.

The possibility needs to be explored where newly developed nosodes could be used on the basis of their known microbiological effects and pathogenesis, without conducting the HPT, as the most important objective of any HPT is to understand the effects on humans, which are already known. If the system of the HPT is suitably supplanted by thorough evaluation of data derived from known microbiological effects, *in vitro* studies, clinical trials

and toxicological studies of nosodes (and other new drug substances), it is likely to facilitate and encourage new drug discovery in Homoeopathy.

CONCLUSION

A need is felt to revamp the old nosodes with newer strains using modern technology and standardization and preparation of both old and new nosodes. It is recommended that nosodes be prepared using fresh source and with appropriate guidelines and documentation to ensure reproducibility and future research. The standardization method of preparation of nosodes will be reproducible if the guidelines laid down are followed by the pharmacies.

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होम्योपैथिक नोसोड को तैयार करने का वैज्ञानिक तरीका

सारः

पृष्ठभूमिः नोसोड होम्योपैथिक औषधि है जिसका स्त्रोत जैविक सामग्री जैसे कि रोगयुक्त ऊतक, जीव सम्बर्धन (जीवाणु, कवक और विषाणु) या परजीवी या पशुओ व मानवो के क्षरित पदार्थ है। सन 1830 से लगभग 45 नोसोड प्रयोग में लाये जा रहे है जिनकी तैयार करने की विधि में सुधार की आवश्यकता है।

उद्देश्यः विभिन्न प्रकार की प्रजातियों के प्रयोग द्वारा नोसोड की बनाने की विधि व मानकीकरण। आधुनिक तकनीक व मानकीकरण द्वारा पुराने नोसोड को, नयी प्रजातियो का प्रयोग कर उच्च स्तर का बनाना। नोसोड श्रेणी में नयी औषधि की खोज के लिये स्पष्ट रूप रेखा तैयार करना।

सामग्री एवं विधियाँ

जैविक पदार्थो की पहचान जैसे कि सम्बर्धन की पहचान, वांछित पदार्थ का शुद्ध, पृथक्करण, मात्रकीकरण, मानकीकरण, तनुता, पोटेन्टाइजेशन विधि एवं नोसोड का सुरक्षित उपयोग समझाया गया व पत्रावलित सुव्यवस्थित तरीके से किया गया है।

परिणामः एचआईवी, एचसीवी, माइकोबैक्टीरियम ट्यूबरकुलोसिस नोसोड को इस पत्र में वर्णित, संशोधित तरीको से तैयार किया गया।