remel

NAC (50 and 100)

INTENDED USE
Remel NAC (50 and 100) is recommended for use in qualitative procedures as a mucolytic agent in the digestion and decontamination of clinical specimens for isolation of acid-fast bacilli and fungi.

SUMMARY AND EXPLANATION
The majority of clinical specimens submitted to the microbiology laboratory for the recovery of acid-fast bacilli are contaminated with more rapidly growing commensal microbial flora. Also, respiratory specimens, such as sputum, often contain mucin which may trap organisms. Such specimens require liquefaction to release the mycobacterial cells prior to the inoculation of media.[1] The N-acetyl-L-cysteine (NAC) method described by Kubica is the most widely used procedure for digestion and decontamination of clinical specimens.[2] In further testing, Reep and Kaplan demonstrated NAC could be used with sodium citrate as a diluent to liquefy specimens for the recovery of fungi.[3,4]

PRINCIPLE
N-acetyl-L-cysteine serves as a mucolytic agent by disrupting disulfide bonds in the mucous of sputum, gastric lavage, and other body fluids.

REAGENTS
NAC 50:
N-acetyl-L-cysteine (CAS 616-91-1) ..... 0.25 g

NAC 100:
N-acetyl-L-cysteine (CAS 616-91-1) ..... 0.50 g

PRECAUTIONS
This product is for *In Vitro* diagnostic use and should be used by properly trained individuals. Precautions should be taken against the dangers of microbiological hazards by properly sterilizing specimens, containers, and media after use. Directions should be read and followed carefully.

STORAGE
Store product in its original container at 2-8°C until used. After reconstitution, NAC (50 and 100) may be stored at 2-8°C for 24 hours. Allow product to equilibrate to room temperature before use.

PRODUCT DETERIORATION
This product should not be used if (1) there is evidence of contamination, (2) the color has changed, (3) the expiration date has passed, or (4) there are other signs of deterioration.

SPECIMEN COLLECTION, STORAGE, TRANSPORT
Specimens should be collected and handled following recommended guidelines.[5,6]

MATERIALS REQUIRED BUT NOT SUPPLIED
(1) Loop sterilization device, (2) Inoculating loop, swabs, collection containers, (3) Incubators, alternative environmental systems, (4) Supplemental media, (5) Quality control organisms, (6) Mycobacteriological safety equipment, (7) Disinfectant, (8) 50 ml, sterile, graduated, plastic, centrifuge tubes, (9) Pipettes, (10) TB Base Digestant (REF R21268) or 2.94% Sodium Citrate (REF R21262), (11) Phosphate Buffer M/15 (pH 6.8) (REF R21256), (12) 10% KOH (REF R21230), (13) Sterile demineralized water, (14) Vortex mixer, centrifuge.

PROCEDURE
Follow established laboratory safety procedures when working with acid-fast cultures and specimens. Consult appropriate references when necessary for detailed procedural information on specimen processing and media inoculation.[5,6]

Mycobacteriological Culture
Reagent Preparation:
NAC (50 and 100) - Use the Ampule safety snap provided to carefully break off the top of the ampule. Add 1 ampule of NAC 50 (REF R21076) to 50 ml of TB Base Digestant (REF R21268) or 1 ampule of NAC 100 (REF R21079) to 100 ml of TB Base Digestant. Swirl to dissolve. The final concentration of NaOH in the tube is 1%.[1]

Specimen Processing:
1. Transfer 10 ml or less of specimen to a sterile centrifuge tube and add an equal volume of NAC/digestant solution.
2. Tighten the cap and invert the tube ensuring that the solution contacts all inside surfaces of the tube and cap.
3. Mix the contents for approximately 20 seconds on a Vortex mixer.
4. Allow the mixture to stand at room temperature for 15 minutes. Specimens should remain in contact with the decontaminating agent for only 15 minutes. Overprocessing may result in reduced recovery of mycobacteria.[1]
5. Swirl the tube periodically to assist in mucolytic action.
6. Add Phosphate Buffer M/15 (pH 6.8) to the 50 ml mark.
7. Recap the tube tightly and invert several times to mix the contents.
8. Place tube in an aerosol-free, sealed centrifuge cup. Centrifuge at ≥ 3000 x g for 15 to 20 minutes.
9. Pour off the supernatant into a splash-proof discard container filled with a suitable disinfectant. Do not allow the disinfectant to flow into the tube. Swab the lip of the tube with disinfectant and recap.
10. Resuspend the sediment (pellet), adding a small amount of Phosphate Buffer if necessary.

11. Use the sediment to prepare stains and cultures.
12. Refer to Clinical Microbiology Procedures Handbook or the CDC manual for further instructions in the digestion and decontamination process and recommended guidelines for processing other specimen types (e.g., gastric lavage, laryngeal swabs, tissue, blood, and other body fluids).[1,5,6]

Mycological Cultures for Sputum[6]

Reagent Preparation:

NAC (50 and 100) – Use the ampule safety snap provided to carefully break off the top of the ampule. Add 1 ampule of NAC 50 (REF 21076) to 50 ml of 2.94% Sodium Citrate (REF 21262) or 1 ampule of NAC 100 (REF 21079) to 100 ml of 2.94% Sodium Citrate. Swirl to dissolve.

Specimen Processing:

1. Transfer 10 ml or less of specimen to a sterile centrifuge tube and add an equal volume of the NAC/digestant solution.
2. Tighten the cap and invert the tube ensuring that the solution contacts all inside surfaces of the tube and cap.
3. Mix each tube on a Vortex mixer until the specimen is liquefied, or almost liquefied, if culturing for *Pneumocystis carinii*. Consult appropriate references for procedures for specific fungi.[5,6]
4. Dilute the solution to the 50 ml mark with sterile demineralized water or Phosphate Buffer M/15 (pH 6.8). Tighten the cap and mix by swirling or inversion.
5. Place the tube in an aerosol-free, centrifuge cup. Centrifuge at 2100 x g for 15 minutes.
6. Pour off the supernatant into a splash-proof discard container filled with a suitable disinfectant. Do not allow the disinfectant to flow into the tube. Swab the lip of the tube with disinfectant and recap.
7. Mix to resuspend the sediment.
8. Use 0.1 ml of sediment to inoculate culture media. Examine a portion of the sediment microscopically in 10% KOH. Other smears may be maintained for future staining.
9. Incubate cultures at 30°C for 4 weeks or longer.

QUALITY CONTROL

All lot numbers of NAC (50 and 100) have been evaluated and found to be acceptable. Testing of control organisms should be performed in accordance with the quality control procedures established by each laboratory following their state and local regulatory agencies. If aberrant quality control results are noted, patient results should not be reported.

BIBLIOGRAPHY

1. Kent, P.T. and G.P. Kubica. 1985. Public Health Mycobacteriology, A Guide for the Level III Laboratory. U.S. Dept. of H.H.S., CDC, Atlanta, GA.
2. Kubica, G.P., A.J. Kaufmann, and W.P. Dye. 1963. Amer. Rev. Resp. Dis. 89:284-286.
3. Reep, B.R. and W. Kaplan. 1972. Health Lab. Sci. 9:118-124.
4. Reep, B.R. and W. Kaplan. 1972. Mycopathol. Mycol. Appl. 46:325-334.
5. Murray, P.R., E.J. Baron, J.H. Jorgensen, M.L. Landry, and M.A. Pfaller. 2007. Manual of Clinical Microbiology. 9th ed. ASM Press, Washington, D.C.
6. Isenberg, H.D. 2004. Clinical Microbiology Procedures Handbook. 2nd ed. ASM, Washington, D.C.
7. McGinnis, M.R. 1980. Laboratory Handbook of Medical Mycology. Academic Press, New York, NY.

PACKAGING

REF R21076, 50 mg/Ampule..... 6/Pk
REF R21079, 100 mg/Ampule..... 6/Pk

Symbol Legend

REF	Catalog Number
IVD	In Vitro Diagnostic Medical Device
LAB	For Laboratory Use
	Consult Instructions for Use (IFU)
	Temperature Limitation (Storage Temp.)
LOT	Batch Code (Lot Number)
	Use By (Expiration Date)

CAS (Chemical Abstracts Service Registry)

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