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A Critical Appraisal of the Utility of the Serum Anticholinergic Activity Assay in Research and Clinical Practice

By Ryan M. Carnahan, PharmD,
Brian C. Lund, PharmD, Paul J. Perry, PhD,
and Bruce G. Pollock, MD, PhD

ABSTRACT ~ The serum anticholinergic activity (SAA) assay was originally designed to quantify the anticholinergic burden of drug exposure. The same assay has been used to measure the anticholinergic activity of standard drug solutions. There are limitations to the use of the assay in research and in applying these findings to clinical practice. Assays of standard drug solutions do not account for pharmacokinetic differences among drugs, which limits the interpretation of such measurements. In addition, emerging evidence has suggested that anticholinergic medications may not be the only cause of elevated SAA. Despite these limitations, elevated SAA has been consistently associated with cognitive impairment and delirium in a number of research settings. Such findings have prompted investigators to consider the potential application of the SAA assay in research and in clinical practice. Therefore, the objectives of this review are to summarize the current literature involving the SAA assay, describe the relative merits and shortfalls of the SAA assay as a research tool, and discuss the potential for use of the SAA assay as a clinical tool. *Psychopharmacology Bulletin*. 2002;36(2):24-39

The Anticholinergic Activity Assay

The anticholinergic activity assay was first described by Tune and Coyle.¹ The assay is performed by incubating a small amount of sample solution in a phosphate buffer containing [³H]quinuclidinyl benzilate (QNB), a potent muscarinic antagonist, and a suspension of rat striatal membranes, which are rich in muscarinic receptors. Antimuscarinic substances in the sample competitively inhibit the bind-

Dr. Carnahan is postdoctoral fellow in clinical psychopharmacology and Dr. Perry is professor in the Clinical and Administrative Pharmacy Division of the College of Pharmacy at the University of Iowa in Iowa City. Dr. Perry is also professor in the Department of Psychiatry of the College of Medicine at the University of Iowa. Dr. Lund is postdoctoral research scholar in pharmacoeconomics at the College of Public Health at the University of Iowa. Dr. Pollock is professor in the Academic Division of Geriatrics and Neuropsychiatry of the Western Psychiatric Institute and Clinic at the University of Pittsburgh in Pennsylvania.

To whom correspondence should be addressed: Paul J. Perry, PhD, University of Iowa, College of Pharmacy, 415 S Pharmacy Building, Iowa City, Iowa 52242-1112; Tel: (319) 335-8803; Fax: (319) 353-5646; paul-perry@uiowa.edu

ing of the radioactively labeled QNB to the receptors to a degree determined by their concentrations and affinity for these receptors. Isolation of the receptor- ^3H -ligand complexes is achieved by filtration over glass fiber filters. The complexes are then counted by scintillation spectrometry to determine the antimuscarinic activity of the sample solution. The spectrometry result is standardized by comparing the binding exhibited by the sample to that of a standard solution, generally a 10^{-8} M solution of atropine, a potent anticholinergic agent. The antimuscarinic activity of the sample is then expressed in "atropine equivalents." One potentially problematic issue that has not been sufficiently addressed is the inability of the assay to differentiate agonists from antagonists. The assay defines anticholinergic activity as the amount of QNB displaced from muscarinic receptors by a sample, relative to the amount displaced by a standard solution of atropine. However, QNB is displaced by both cholinergic agonists and antagonists.

The anticholinergic activity assay has been used in two primary applications, which differ by the sample solution employed. These applications are the measurement of anticholinergic activity in standard drug solutions and in patient serum (ie, serum anticholinergic activity [SAA]).

Anticholinergic Activity in Standard Drug Solutions

Tricyclic antidepressants, antipsychotics, antihistamines, cycloplegics, antispasmodics, antiparkinsonian agents, proprietary sleeping medications, belladonna alkaloids, and some toxic plants are among the compounds that have long been known to have anticholinergic effects. More recently, it has been demonstrated that many medications not generally considered anticholinergic may actually have some anticholinergic properties. Tune et al² quantified the anticholinergic activity of the top 25 medications prescribed to the elderly, according to a Health Care Financing Administration listing from the late 1980s. The anticholinergic activity of a standard concentration (10^{-8} M) of each compound was assayed using the radioreceptor methodology, as previously described. Measurable anticholinergic activity was observed with 13 of the studied drugs: captopril, cimetidine, codeine, digoxin, dipyridamole, furosemide, isosorbide dinitrate, nifedipine, theophylline, tiamterene/hydrochlorothiazide (hydrochlorothiazide alone did not produce detectable levels), prednisolone, ranitidine, and warfarin. Researchers have continued to test medications' anticholinergic activity, but only some of these results are available in the literature.³

Although the basic concept is good, one must use caution when interpreting the anticholinergic activity of standard drug solutions. The assay attempts to compare two drugs and concludes that the drug with the

higher atropine equivalent measurement is “more anticholinergic.” However, the magnitude of anticholinergic activity cannot be reliably compared across drugs because the assay uses the same standard concentration for each drug (10^{-8} M), which does not necessarily reflect any biologically meaningful serum concentrations. As a result of differences in dose, absorption, distribution, protein binding, production of active metabolites, and elimination rate (ie, pharmacokinetics), drugs simply do not appear at the same molar concentrations in the serum. These differences are further complicated by the relative degree of penetration into the central nervous system (CNS), which is presumably the site where anticholinergic-related changes in mental status are produced. In addition, muscarinic receptor subtypes differ in their central effects, some enhancing and some inhibiting various aspects of neurotransmission, but the assay is not specific to receptor subtype. It is also tempting to sum atropine equivalents for individual drugs across a patient’s drug profile to come up with some summary measure of anticholinergic exposure. Such inferential applications share the same pharmacokinetic shortcomings as between-drug comparisons. In short, the anticholinergic activity of standard drug solutions is likely not a reliable measurement of the magnitude of anticholinergic activity. However, the qualitative recognition of subtle anticholinergic properties associated with drugs not generally considered anticholinergic (eg, digoxin, warfarin, etc.) is clinically relevant. Although none of these drugs is likely to produce delirium alone, the combination of several agents may predispose patients to developing delirium, if, in fact, assay measurements do reflect anticholinergic properties.

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Anticholinergic Activity in the Serum

Researchers have found associations between elevated SAA and mental status changes in a number of clinical settings and patient populations. The degree of mental status change investigated varies across studies, but falls into two general categories: (1) the association between SAA and delirium, and (2) the association between SAA and subtle cognitive changes. The principal findings from studies using the SAA assay are briefly discussed in the following sections. These summaries will provide the basis for an assessment of the utility of the SAA assay.

Serum Anticholinergic Activity and Delirium

Delirium is an acute state of cognitive impairment that may involve disturbances in perception, emotion, awareness, memory, thinking, orientation, the sleep-wake cycle, and psychomotor behavior. These disturbances may vary in severity and presentation throughout the course of a day.⁴ The majority of delirious patients have serious medical, surgical, or

neurological illnesses or are in a state of drug intoxication or withdrawal.⁵ It is estimated that 10% of patients over 65 years of age who are hospitalized due to a general medical condition are delirious on admission, and an additional 10% to 15% may become delirious while in the hospital.⁴ The incidence of delirium increases with the number of risk factors.⁶ The estimates of delirium incidence and prevalence vary across studies and patient populations. One study of long-term care facilities found 40.5% of residents (15/37) to be delirious at some time during the 2-week observation period.⁷ Delirium impacts not only the individual but the healthcare system as well. Increased morbidity and mortality, functional decline, increased length of hospitalization, and requirements for additional care are associated with delirium.⁸⁻¹²

Anticholinergic medications and substances are a well-known cause of delirium. This is presumably due to a direct reduction in central cholinergic activity and the resultant changes in neurotransmission. Numerous challenge studies have found impairments in various aspects of cognitive function after administering standard therapeutic doses of anticholinergic medications to normal healthy adults, regardless of age.¹³⁻¹⁶ It is clear that anticholinergic medications can have a negative impact on cognition. The association between SAA and the risk of delirium has been examined in several clinical studies.

Tune et al¹⁷ examined the SAA levels of 25 postoperative cardiac patients from 29–75 years of age, who were evaluated for delirium. An SAA threshold of 7.5 pmol/mL atropine equivalents was significantly associated with an increased risk of delirium. Seven of the eight delirious patients had SAA levels above this threshold, compared to only 4 of 17 nondelirious patients ($P=.001$). Furthermore, reductions in scores on the Mini-Mental State examination¹⁸ (MMSE) correlated with SAA levels ($r=0.83$; $P=.001$).

Golinger et al¹⁹ studied the association of SAA and delirium in 25 surgical patients in the intensive care unit ranging from 25 to 76 years of age. Most patients had undergone a major surgical operation, except for two who were being treated nonsurgically for inflammatory conditions. Patients were categorized as delirious or nondelirious based on *Diagnostic and Statistical Manual of Mental Disorders*,²⁰ Third Edition, criteria for delirium, and a serum sample for the SAA assay was drawn within 4 hours of the mental status examination. Mean SAA was significantly greater in delirious patients than in nondelirious patients (4.67 and 0.81 ng/mL atropine equivalents, respectively; $P=.007$).

Mach et al²¹ measured SAA in 11 delirious but nondemented male patients over 60 years of age in comparison to 11 age-matched controls. All were inpatients on medical wards. At baseline, the mean

SAA level was higher in the delirious group than in controls ($P < .05$). Six of the patients with delirium had resolution of symptoms before leaving the hospital or extended care ward. These patients actually had higher baseline SAA levels than those whose delirium did not resolve ($P < .05$). In patients whose delirium resolved, mean SAA was also significantly lower after symptom resolution than during delirium. It is difficult to interpret these findings because of the many factors that may influence delirium, but each of the patients whose delirium resolved had at least one medication discontinued between baseline and resolution. Some of these medications had known anticholinergic activity, and some did not. This may reflect a lack of data regarding the anticholinergic activity of some medications, inhibition of medication metabolism during delirium, or, possibly, a reduction in unknown endogenous anticholinergic substances.

Flacker et al²² found higher SAA levels to be independently associated with delirium in 67 consecutively admitted medical inpatients over 75 years of age ($P = .006$). Using the Confusion Assessment Method²³ (CAM) and the Delirium Symptom Interview²⁴ (DSI), 20 patients (30%) met the criteria for delirium. The patients were further divided into quintiles based on their SAA levels, with quintile 1 having the lowest SAA levels and quintile 5 having the highest. The prevalence of delirium increased steadily from 7.7% in quintile 1 to 61.5% in quintile 5.

Mussi et al²⁵ studied risk factors for delirium in 61 patients consecutively admitted to a geriatric medical ward. SAA samples were drawn on admission. Administration of the CAM within 24 hours of admission identified 12 patients with delirium. Among multiple potential risk factors, antipsychotic use, benzodiazepine use, and elevated levels of SAA were independently associated with the presence of delirium ($P < .002$, $P < .005$, and $P < .004$, respectively). The mean SAA was 23.0 pmol/mL atropine equivalents in the delirious group and 3.9 pmol/mL in the nondelirious group. All patients with SAA greater than 20.0 pmol/mL were delirious. It is difficult to determine with certainty whether elevated SAA was a specific cause for delirium, rather than a marker or effect of other causes, but an association was clearly present. The role of antipsychotics and benzodiazepines is unclear since these may be surrogate markers for agitation or anxiety preceding delirium, or they may have been used to treat symptoms of delirium.

Flacker and Lipsitz²⁶ measured SAA in 22 nursing home residents in the midst of febrile illnesses, then again at a 1-month follow-up. Illnesses were not severe enough to require hospitalization. Subjects were not more than moderately demented, as determined by scores of

0–3 on the Cognitive Performance scale²⁷ (CPS; possible scores range from 0 to 6 with higher scores indicating more impairment). Data collected also included the MMSE and DSI at baseline and follow-up. The Cumulative Illness Rating Scale²⁸ (CIRS) was used to determine comorbidity at the time of enrollment. At baseline, eight patients were identified as delirious using the CAM. Those in the delirious group had higher CPS scores than did the nondelirious group ($P<.01$). The delirious group also tended to have higher CIRS scores ($P<.09$), indicating a trend toward greater comorbidity. Age, gender, body mass index, and number of medications did not differ between the groups. SAA was not significantly different between the groups at baseline or follow-up. In the delirious group seven of the eight patients showed an absolute decline in SAA from baseline to follow-up, as did 13 of 14 patients in the nondelirious group. Interestingly, the only patient who was still delirious on follow-up was the one whose SAA had not decreased. The authors speculated that the lack of differences in SAA between the groups could have reflected an increased sensitivity to anticholinergic activity in the brains of patients who were previously cognitively impaired, or that other causes of delirium could have been present. Importantly, the decline in SAA did not appear to be related to medication changes. This observation suggests that either (1) the metabolism of medications was inhibited during illness, raising SAA; or (2) there was another endogenous source of SAA during illness.

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Serum Anticholinergic Activity and Cognitive Changes

Rovner et al²⁹ studied the relationship between SAA levels and self-care capacity in 22 demented nursing home patients, using the self-care subscale of the Psychogenic Dependency Rating Scale.³⁰ This scale specifically measures impairments in dressing, personal hygiene, mobility, toileting, and urinary continence. Observed SAA values ranged from 0–9.95 pmol/mL atropine equivalents with a median of 0.83. Patients with levels above the median SAA displayed significantly greater impairment in self care than did patients below the median ($P=.03$). These groups did not differ with respect to type of dementia, prevalence of arthritis, use of antipsychotics, age, or sex.

Miller et al³¹ evaluated cognitive function in relation to SAA in 36 presurgical patients from 59 to 81 years of age (mean=67 years) in a randomized, double-blind trial. Half of the patients received an intramuscular injection of scopolamine, an anticholinergic agent, 2 hours before surgery, and half received placebo. The scopolamine dose was 0.005 mg/kg, except in the 81-year old patient, who received 0.0025 mg/kg. Cognitive testing was performed the night

prior to surgery and again 45 minutes to 1 hour after the injection. SAA was measured on the evening of baseline testing and again from blood drawn at induction of anesthesia. Analyses of covariance were used to control for baseline differences in cognitive testing in order to determine if the postinjection differences were truly significant. Postinjection testing found no differences in MMSE total or serial sevens scores or in Symbol Digit Modalities Test³² scores between the groups. However, the scopolamine group did recall fewer words than the placebo group on the fifth trial of the Rey Auditory Verbal Learning Test³³ (RAVLT; $P < .01$). In this test, 15 words are read to the patient for a total of seven trials, and recall is tested between each trial. The sixth trial uses a distracter list of different words. The placebo group had numerically superior scores on each trial, but differences were not significant. SDS scores showed that there were more delirium symptoms in the scopolamine group than in the placebo group ($P = .02$). The postinjection SAA levels also differed, with means (\pm SDs) of 121.1 ± 85.5 and 11.6 ± 18.2 pmol/mL atropine equivalents in the scopolamine and placebo groups, respectively ($P = .0001$), indicating a drug effect on SAA. This is compared to a baseline SAA of 9.1 pmol/mL atropine equivalents for both groups combined. Patients were also grouped according to "negligible" or "low" SAA (< 45 and 45 – 200 pmol/mL atropine equivalents, respectively), the cutoff being two SDs above the mean for the overall sample at baseline. The group with "low" SAA levels scored worse on the fifth trial of the RAVLT than did the "negligible" SAA group ($P < .025$). They also performed worse on the serial sevens section of the MMSE ($P < .05$).

It is difficult to determine the clinical significance of these findings, because differences between groups were only found on a minority of the cognitive tests. Notably, the magnitude of the postinjection SAA levels, even in the "low" SAA group, was considerably higher than those reported in other studies, even in delirious patients. This may reflect variability in assay technique or methodology across investigators. Alternatively, the elevated SAA levels could also be a true representation of the direct administration of a potent anticholinergic agent. If this is the case, it would show quite clearly that, although SAA may be elevated in delirious patients, grossly elevated SAA levels do not necessarily result in delirium, as other studies may have suggested. One of the more interesting findings of this study was related to the assessment of anticholinergic activity from cerebrospinal fluid (CSF) in nine patients receiving spinal anesthesia. The anticholinergic activity measured in the CSF correlated significantly with the anticholinergic activity measured in the serum ($P < .05$). However, none of the patients in the placebo

group had detectable CSF anticholinergic activity, despite detectable SAA levels. This novel finding suggests that the presence of measurable anticholinergic activity in the serum does not necessarily translate into measurable CNS exposure. The correlation between CSF anticholinergic activity and SAA is likely due to scopolamine freely crossing the blood brain barrier in proportion to blood levels. Other drugs, however, may contribute to anticholinergic activity measured peripherally in the serum, but do not necessarily penetrate the CNS and exert central anticholinergic activity and toxicity.

Thienhaus et al³⁴ studied the effects of increased SAA in 10 patients with probable Alzheimer's dementia in comparison with 18 patients not exhibiting cognitive impairment. All subjects were inpatients on a geropsychiatric service. Baseline assessments of cognition and SAA were performed prior to the initiation of whatever antidepressant or antipsychotic treatment was clinically indicated. The nondemented group had scores of at least 25 on the MMSE and primarily consisted of patients with diagnoses of major depressive disorder, bipolar disorder, or schizophrenia. Most patients had either no prescribed medications or had been medication noncompliant. Antiparkinsonian agents were used as needed to control extrapyramidal side effects. Any other prescribed medications were kept at constant dosages. Follow-up assessments were performed after at least 1 week of a stable medication regimen. The average SAA in the demented group increased from a baseline of 3.5 to 6.17 nmol/L atropine equivalents after the implementation of drug therapy ($P < .01$). This change was associated with significant worsening on a number of cognitive measurement scales. In the nondemented group, the mean SAA changed significantly from 4.09 to 6.66 nmol/L atropine equivalents. Unlike in the demented group, however, no cognitive impairment was observed, despite an almost identical change in SAA. Thus, despite the heterogeneity of psychiatric illnesses in the nondemented group and the potential for confounding by these illnesses, this study suggests that demented patients may be more susceptible to the detrimental cognitive effects of anticholinergic medications than nondemented patients. It is also notable that both groups had detectable levels of SAA at baseline, despite the fact that most patients were reportedly not taking medications.

Tollefson et al³⁵ studied the effects of reducing anticholinergic medications on mental status and SAA. The study population was 34 residents from three different nursing homes who had received at least one anticholinergic medication at a stable dosage for at least 2 weeks and were over 65 years of age (mean=79 years). Exclusion criteria included

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acute medical or psychiatric illness that could adversely affect cognition, medications that were only given “as needed,” presence of “non-anticholinergic” medications that could adversely affect cognition, significant dementia or delirium, physical problems interfering with psychometric testing, and inability to provide informed consent. Baseline cognitive testing was performed, and SAA was measured. The goal of the intervention was to reduce the patient’s anticholinergic burden by 25%, based on an “anticholinergic index” score derived from the patient’s treatment regimen.³⁶ Patients were randomized to control and intervention groups. The intervention was successfully achieved in 15 patients (2 dropped out after deterioration). The mean (\pm SD) SAA at baseline was 2.49 ± 3.9 ng/mL in the intervention group and 3.58 ± 3.8 ng/mL in the control group. One month after the intervention, the mean SAA was 1.89 ± 3.4 ng/mL in the intervention group and 3.23 ± 3.7 ng/mL in the control group. Attempts were made to correlate the changes in scores on 15 cognitive tests with the magnitude of changes in SAA. The Saskatoon Delirium Checklist³¹ was the only test on which change scores approached significance ($P=.06$). Although the statistical analysis was not presented, it also does not appear that the magnitude of change in mean SAA was significantly different between the intervention and control groups.

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Nebes et al³⁷ studied the relationship between SAA and verbal memory performance in 36 geriatric patients diagnosed with major depressive disorder. Patients with a clinical diagnosis of dementia or a score of less than 126 on the dementia rating scale³⁸ (DRS) were excluded. The Cumulative Illness Rating Scale-Geriatric³⁹ (CIRS-G) was used to obtain a global measure of the severity of medical conditions. Patients were divided into two groups, one with detectable SAA ($n=19$) and the other without ($n=17$). These groups did not differ significantly with respect to baseline CIRS-G or DRS scores. Age and Hamilton Rating Scale for Depression⁴⁰ scores at baseline were significantly greater in the group with detectable activity, however, and were therefore controlled for in the comparison of cognitive testing between groups. The group with detectable levels of SAA showed more impairment in delayed recall ($P<.05$) and percent retention ($P<.05$). These memory parameters reflect both the ability to recall a list of 15 words 30 minutes after four immediate recall trials as well as immediate recall abilities throughout these trials. Significant deficits were seen despite a mean SAA of less than 0.3 pmol/mL atropine equivalents and no subject having a level above 1.0 pmol/mL. The authors contrasted this with other studies in which higher levels of SAA correlated with memory impairment, and studies in which even high levels did not correlate with impairment.

They suggested that old age and/or depression in their patients may have increased the patients' sensitivity to anticholinergic effects. Although the memory impairments in this study were mild, it does suggest that even small amounts of anticholinergic activity may be sufficient to inhibit cognition in some patients.

Utility of the Serum Anticholinergic Activity Assay in Clinical Research

Numerous studies have used the SAA assay to study mental status changes, ranging from subtle cognitive changes to delirium. What information has been gained from these studies regarding the utility of the SAA assay as a clinical research tool? One issue relates to whether the assay is able to measure known changes in anticholinergic status. For example, the administration of anticholinergic drugs should produce an increase in SAA if the assay is a valid and useful measure of anticholinergic activity. At least two studies have measured SAA prior to and after the initiation of anticholinergic drugs and observed significant increases in SAA.^{31,34} A third study demonstrated an apparent decrease in SAA after the reduction of anticholinergic medications, though this decrease did not appear different from the group without medication changes.³⁵ A related concern is whether anticholinergic activity measured in the serum accurately reflects anticholinergic activity in the CNS. The one study that reported both serum and CSF anticholinergic activity had somewhat conflicting findings.³⁹ Globally, however, the fact that so many studies have positively correlated SAA levels with delirium and cognitive impairment supports the SAA assay as a reasonable marker for central anticholinergic activity.

A second issue relates to whether the tool has some meaningful relationship to the clinical construct being measured. That is, does SAA correlate with the degree of cognitive impairment, or does the assay differentiate patients with and without delirium? In the majority of studies conducted with the SAA assay, some association along these lines was established. The strongest evidence was provided by Flacker and colleagues,⁴¹ who observed a clear increase in delirium risk across increasing quintiles of SAA. This is analogous to a dose-response relationship, which greatly strengthens the evidence of association between SAA and delirium.

Unfortunately, the precise nature of this association has been difficult to characterize because of the heterogeneous methodologies used across studies. In the case of delirium, one study identified a threshold level of SAA that was associated with increased risk.¹⁷ Three other studies reported higher mean SAA levels in delirious versus nondelirious

patients, but did not report any particular threshold for increased risk.^{19,21,24} In the case of cognitive impairment, investigators compared groups with detectable versus undetectable SAA,³⁷ groups with SAA levels above versus below a median value,²⁹ and cognitive changes seen after instituting^{31,34} or discontinuing³⁵ drugs. An additional barrier to synthesizing these studies is that the units of SAA have been expressed in at least five different ways, including pmol/mL, ng/mL, nmol/L, μ M, and nM(/200 μ L). Even after converting units, there are disparities between studies in the magnitude of SAA that is associated with a particular outcome, such as delirium (Table). The one study³⁴ reporting results as μ M concentrations had values differing from most others by a factor of 1,000. Studies also indicate that certain populations (ie, elderly, demented) are impaired at lower levels of SAA than are others, perhaps reflecting a relative deficit of "cholinergic reserve" in these populations. Unfortunately, given the state of the current literature, the

TABLE

RANGE OF SAA LEVELS ACROSS STUDIES

Study (Population)

GROUP	MEAN AGE (years \pm SD)	MEAN SAA*	MEAN CONVERTED SAA* (pmol/mL)
<i>Nebes et al³⁷ (Depressed geriatric patients)</i>			
SAA detectable	70.9 \pm 6.7	0.28 pmol/mL	0.28
SAA undetectable	67.2 \pm 5.8	0 pmol/mL	0
<i>Theinhaus et al³⁴ (Elderly psychiatric patients)</i>			
Alzheimer's disease	66 \pm 11	3.50 μ M	3,500
Nondemented	64 \pm 8	4.09 μ M	4,090
<i>Rovner et al²⁹ (Demented nursing home patients)</i>			
Divided into groups above and below median SAA	80.8 \pm 9.6	Mean not reported. Median=0.83 pmol/mL; range=0–9.95 pmol/mL	0.83 (median)
<i>Tollefson et al³⁵ (Elderly nursing home patients)</i>			
Control	For all subjects:	3.58 ng/mL	5.15
Intervention	79.0 \pm 9.7	2.49 ng/mL	3.58
<i>Mussi et al²⁵ (Geriatric medical patients)</i>			
Delirious	77.5 \pm 10.1	23.0 pmol/mL	23.0
Not delirious	79.9 \pm 12.2	3.9 pmol/mL	3.9
<i>Golinger et al¹⁹ (Surgical ICU patients)</i>			
Delirious	60; range=29–74	4.67 ng/mL	6.72
Not delirious	57; range=29–76	0.81 ng/mL	1.17

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SAA assay cannot be used to predict risk of delirium or other cognitive impairment in an individual patient.

Despite heterogeneity in methodology and magnitude of SAA results, the fact remains that most studies have found a relationship between SAA and mental status changes. Arising from this observation is a more fundamental question: what exactly is the SAA assay measuring? One potential use of the SAA assay is to assess overall anticholinergic burden from medications. As discussed in the section regarding anticholinergic activity in standard solutions, accurately determining the relative in vitro anticholinergic potency of various drugs and drug combinations is extraordinarily difficult. Measuring SAA would conceptually eliminate the need to make these determinations by directly measuring the extent of anticholinergic burden. Thus, researchers interested in studying or controlling for the effect of

TABLE (CONTINUED)

RANGE OF SAA LEVELS ACROSS STUDIES

Study (Population)

GROUP	MEAN AGE (years±SD)	MEAN SAA*	MEAN CONVERTED SAA* (pmol/mL)
<i>Tune et al</i> ¹⁷ (Postcardiac operation patients)			
Delirious	For all subjects: 55; range=29–75.	Mean not reported. 7/8 patients >7.5 pmol/mL	
Not delirious		Mean not reported. 13/17 patients <7.5 pmol/mL	
<i>Flacker et al</i> ²² (Medical inpatients >75 years)			
Delirious	86.2±6.5	1.8 nM (/200 µL)	1.8
Not delirious	85.2±6.0	0.7 nM (/200 µL)	0.7
<i>Flacker and Lipsitz</i> ²⁶ (Nursing home patients with febrile illness)			
Delirious	For all subjects:	0.69 nM (/200 µL)	0.69
Not delirious	88.0±4.5	0.65 nM (/200 µL)	0.65
<i>Mach et al</i> ²¹ (Patients >60 years and not demented)			
Delirious	71.8±8.0	6.05 nM	6.05
Not delirious	70.8±6.0	3.38 nM	3.38
<i>Miller et al</i> ³¹ (Presurgical patients 59–81 years)			
Not separated at baseline	67.0±5.9	9.1 pmol/mL	9.1
<i>Flacker and Wei</i> ⁴¹ (Elderly medical inpatients)			
8/10 patients had detectable SAA	Mean not reported. All subjects >70	0.69 nmol/L	0.69

*Mean values at first measurement.

SAA=serum anticholinergic activity; ICU=intensive care unit.

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anticholinergic medications on the risk of delirium could theoretically do so with one simple continuous variable, rather than using an imprecise or complex analysis derived from a patient's drug regimen.

Unfortunately, the use of SAA as a measure of anticholinergic burden from medications is confounded and likely impossible to interpret. The first piece of evidence comes from studies of delirium risk factors. These studies often fail to identify anticholinergic medications as a risk factor for delirium, indicating that this is a relatively uncommon cause.^{6,42} However, the delirium studies using SAA have consistently demonstrated an association between anticholinergic activity and delirium. The second piece of evidence comes from studies measuring SAA before and after delirium resolution. Although delirium resolution was accompanied by a decrease in SAA, the change was often not attributable to discontinuation or reduction in anticholinergic medication.^{21,26} The final piece of evidence comes from a recent study that measured SAA in 10 elderly hospital inpatients (>70 years of age) who had not received anticholinergic medications for at least 1 week prior to admission.⁴¹ Eight of the 10 patients had detectable SAA, ranging from 0.23 to 1.72 nmol/L atropine equivalents (mean=0.69 nmol/L). The medications these patients were taking were then assayed for anticholinergic activity at normal therapeutic concentrations. None of these medications registered on the assay as being anticholinergic. Furthermore, one patient was not taking any medications but had detectable SAA.

Considered together, these pieces of evidence indicate that anticholinergic medications are not the only factor that can contribute to SAA and specifically point to the likelihood of endogenous compounds with anticholinergic activity. Implicated compounds include dynorphin A, myelin basic protein, protamine, and cortisol.⁴³⁻⁴⁵ Of particular interest may be cortisol, which is known to increase during stress.⁴⁶ The implication of endogenous compounds also calls into question the cause-effect relationship in the association of SAA and delirium. Does the state of elevated anticholinergic activity actually cause delirium, or is it merely a reflection of endogenous compounds released in response to acute illness? If the former is true, what is causing the elevated state? Could elevated anticholinergic activity be a common pathway for many of the medical causes of delirium, such as infection, metabolic disorders, and so on?

In summary, this comprehensive review of the literature firmly supports the association between SAA and mental status changes, including delirium and cognitive impairment. However, anticholinergic medications are apparently not the only determinant of SAA, and

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investigators are cautioned to give thoughtful consideration to the interpretation of findings using the SAA assay. If elevated SAA is associated with a particular outcome, what does that actually mean? Is elevated SAA potentially caused by the outcome? If SAA is being used as covariate in assessing the relationship between delirium and a given risk factor, what is really being controlled for?

Implications for Clinicians

What can clinicians learn from the research utilizing the SAA assay? First, the association of impaired cognition and delirium with elevated SAA simply reinforces the practice of limiting the use and dosages of medications with anticholinergic activity. In vitro determination of anticholinergic activity suggests that a greater number of medications have anticholinergic activity than were previously thought. This encourages the practice of weighing the risk-benefit ratio of every medication that a patient is taking. If a patient begins to show signs of cognitive impairment, however mild, medications should be analyzed as possible causes and discontinued if appropriate. Although the measured anticholinergic activity of a standard solution of a drug does not necessarily reflect true anticholinergic potency, it is still helpful for clinicians to be aware of which drugs may potentially have anticholinergic effects.

The ultimate question raised after examining these studies is whether the SAA assay could be a useful clinical tool. Elevated SAA has been clearly associated with cognitive impairment and delirium. In patients who are already delirious, the SAA assay has little potential for clinical utility because any nonessential anticholinergic medications should be discontinued anyway. If future research identifies other medical causes of delirium that selectively elevate SAA, the assay may be useful to aid in differential diagnosis and more rapid identification of the underlying cause.

Perhaps the most promising clinical application of the SAA assay would be to prevent delirium by identifying patients at risk. Unfortunately, the current research has not identified consistent levels of SAA that would be useful in identifying patients at risk for delirium. If normal and toxic ranges of SAA could somehow be established by future investigations, then the assay may prove clinically useful. Again, this is complicated by the observation that different populations show impairment at differing levels of SAA. It is evident that a better understanding of the assay itself and what precisely it measures is needed before considering the use of the SAA assay as a clinical tool. ♣

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*Carnahan,
Lund, Perry,
and Pollock*

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