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n-Butyl 2-Cyanoacrylate—Substitute for IBCA in Interventional Neuroradiology: Histopathologic and Polymerization Time Studies

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Despite the development of new alternative embolic agents, the endovascular treatment of brain arteriovenous malformations continues to frequently require the use of cyanoacrylic glue, especially in situations where particulate or sclerosing agents are ineffective, such as when flow is very rapid or fistulous. Because isobutyl 2-cyanoacrylate (IBCA), the most commonly used embolic glue, is no longer available or manufactured, a real need exists for an alternative fast polymerizing agent. In vivo and in vitro studies were performed to compare IBCA with *n*-butyl 2-cyanoacrylate (NBCA, supplied as Histoacryl Blue), a tissue adhesive approved for surgical use in some countries. Polymerization times in static plasma were compared, and the effect of the addition of iophendylate oil or glacial acetic acid on polymerization was assessed. Polymerization times in vivo were compared after intraarterial injection into the internal carotid artery in pigs using a standardized technique. The histopathologic reactions to each glue in the embolized pig rete were assessed and compared over a period of 0–60 days after embolization. Our results show that while NBCA polymerization is demonstrably faster than IBCA in vitro, intraarterial injections of each glue show no significant difference in polymerization times. Like IBCA, NBCA polymerization can be predictably prolonged by the addition of oil or glacial acetic acid, though the effect is less for NBCA. Histopathologic reactions were similar, with acute vasculitis observed, becoming chronic and granulomatous after about 1 month. Both glues showed frequent foci of extravascular extrusion through the embolized rete and recanalization of previously occluded embolized vessels.

We conclude that NBCA has clinical and biologic behaviors similar to IBCA, and therefore should be an acceptable alternative to IBCA for intravascular use.

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The unique properties of cyanoacrylic glue make it sometimes indispensible as an embolic material for the endovascular therapy of arteriovenous malformations (AVMs) [1–6]. Of the various known alkyl-cyanoacrylate homologues, only isobutyl 2-cyanoacrylate (IBCA, bucrylate*) has been used extensively for this purpose and its effects documented [2, 3, 7–9]. In the United States, IBCA use has always been considered experimental, requiring an Investigational Drug and Device Exception from the Food and Drug Administration. Recently, on the basis of an unpublished animal study, the manufacturer discontinued production of IBCA when it was found that liver sarcoma was induced in rats after intraperitoneal injection of large doses of the glue [10–12]. Yet, in many years of cyanoacrylic use in humans involving thousands of patients, not one documented case of carcinogenesis has been reported [10]. Histologic studies of tissue surfaces coated with cyanoacrylic have revealed a wide spectrum of local toxicity, with intensity of reaction inversely proportional to the length of the alkyl side chain [13–17]. Few of the published reports on intravascular use of IBCA in humans and animals [8, 18–23] have described in detail the chronic histopathologic reactions induced in the vessel embolized. The chronic reactions to emboli of other 2-alkyl homologues such as ethyl [24] and *n*-butyl [25–27] have not been analyzed.

As a possible substitute for IBCA, we investigated a chemically similar monomer,

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n-butyl 2-cyanocrylate (NBCA), which has been in surgical use for many years as a tissue adhesive and suture substitute [13–15]. We sought to evaluate the relevant characteristics of NBCA as an embolic material in vitro and in vivo, to determine whether it could be substituted for IBCA, what modifications in technique if any its use would necessitate, and how its histotoxicity compares with that of IBCA on intravascular injection.

Materials and Methods

The form of NBCA studied was Histoacryl Blue,[†] which is supplied in 0.5-ml vials and is colored to increase its visibility in surgical use. As recommended by the manufacturer, Histoacryl was kept in refrigerated storage. The IBCA used was bucrylate. Glues were used at room temperature (approximately 22°C).

In Vitro Studies

To compare polymerization time of the two glues, the following experiments were done. Each glue was drawn up into a 1-cm^3 tuberculin syringe, and a single droplet slowly ejected from a 25-gauge needle was dropped from a standard height onto human plasma at room temperature, by a single investigator. Droplet size was not measured, but appeared consistent during several hundred trials. The plasma was contained in a transparent syringe cap imprinted with newsprint-sized lettering readable through the plasma. The time from contact of glue with plasma to polymerization was measured with a stopwatch by a single observer. Polymerization was defined as the time at which the glue had become sufficiently opaque to render the lettering illegible.

IBCA polymerization can be prolonged by adding small amounts of iophendylate oil [28] (Pantopaque[‡] or Ethiodan[§]) and/or glacial acetic acid [3, 9, 29], creating a condition often clinically useful in AVM embolization [2, 3]. Mixtures of glue and oil used in human embolizations are prepared in proportions commonly ranging from 0:1 (no oil) to 1:1 (oil/glue volume ratios). We usually use a 0.3/1 volume ratio. If desired, additional prolongation is obtained by adding a 10- or 20-μl aliquot of acid/ml glue. These proportions were derived empirically, from clinical observation of polymerization times during human AVM embolizations.

To more fully compare the behaviors of NBCA with IBCA, a wider range of glue/oil and glue/acid mixtures was studied. Graded amounts of iophendylate or glacial acetic acid were added to each glue, and the effect of polymerization time observed. The microliter volumes of acid required were measured with a micropipette.[‖] On average, 20 separate measurements of polymerization times were made for each different mixture and preparation: glue alone, glue plus oil, and glue plus oil and acid.

In clinical use, powdered tantalum is usually added to the glue/oil mixtures [3] to provide radiopacity (1 g tantalum/1 ml glue). This mixture, however, is quite opaque visually due to the black tantalum, precluding an assessment of polymerization time by the above method. Yet, we wanted to examine the effect of tantalum on polymerization, as we had observed a unique "spontaneous" polymerization of NBCA usually within an hour or two after mixing with tantalum and oil. We had not seen this effect with IBCA. Therefore, in a separate experiment, standard mixtures of glue, oil, and tantalum (1 ml, 0.3 ml, and 1 g, respectively) were observed in tuberculin syringes, and the time at which the mixture could no longer be ejected from the syringe was compared for each glue.

In Vivo Studies

The polymerization time of these glues in static plasma is not directly applicable to clinical use as the process is speeded significantly in a flowing system, likely due to increased availability of catalytic anions [29]. Therefore, we performed intraarterial injections of a standard glue/oil mixture in proportions that mimic our clinical preparation. The mixture (1.0 ml glue, 0.3 ml iophendylate, and 1.0 g tantalum) was chosen to minimize the effects of added oil on both polymerization time and tissue pathologic reaction, to better compare the glues themselves.

Domestic swine (20–40 kg) were used. Under general endotracheal halothane anesthesia, percutaneous Seldinger puncture of the right common iliac artery was performed. This technique was nearly always successful and precluded the need for a cutdown. An arterial puncture needle was passed at a 45° angle through a 1-cm skin incision, in a path crossing two fluoroscopic landmarks: the superolateral corner of the acetabulum and the inferior-most edge of the ipsilateral sacroiliac joint. Multiple passes were frequently required. Once entered, a guidewire was positioned in the artery, allowing placement of a 5-French sheath. A tapered-tip 4.1-French catheter then was passed into the origin of an "internal carotid" [30] artery (ICA) under fluoroscopic guidance.

After angiographic assessment of the ICA and rete [30], the catheter was well flushed with dextrose 5% and then 0.15 ml of the glue/oil/tantalum mixture was injected into the catheter dead space. The glue was then "pushed" along by slow hand injection of 5 ml dextrose 5% behind it. The distances each glue bolus traveled in the ICA and rete before hardening into a cast were compared. The rate of injection was kept to a "trickle," as observed fluoroscopically. This was critical for two reasons: the distance a glue bolus traveled could be artifactually increased by more forceful injection, driving it distally; and such distal embolization frequently resulted in stroke or death.

After another dextrose 5% flush (10 ml), the catheter was withdrawn into the common carotid artery. Usually the catheter tip was adherent to the ICA injection site, but was easily detached by gentle traction in all instances. Immediate angiography was then performed through the same catheter, documenting complete occlusion of the ICA in all cases.

Other mixtures were studied in a limited way. In one, acetic acid 20 μl was added to the above glue/oil/tantalum proportions. In another, a 50/50 mixture of glue and oil was used. Each such mixture (0.15 ml) was embolized into the rete of at least two pigs. The prolongation of polymerization that resulted led to unacceptably high morbidity due to cerebral embolization, and precluded further study.

The ICA/rete was unilaterally embolized using these protocols in 33 pigs. It was found through trial and error that extensive rete filling always resulted in a severe neurologic deficit or death due to accompanying cerebral embolization, necessitating immediate sacrifice of the animal. An ideal result was one in which the glue was deposited in the inferior-most aspect of the rete, and not beyond (Fig. 1). Overall, only 18 animals survived neurologically intact, having been unilaterally embolized with the standard embolic mixture (nine for each glue). Animals were sacrificed at intervals of 0–60 days by overdose with IV pentobarbital. The rete structure was then removed and fixed in formalin. A single block was cut through the embolus-containing portion of rete and also from the opposite nonembolized side in each

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Fig. 1.—Gross fresh bilateral rete; inferior aspect of right rete shows black embolus of glue mixture (*arrows*). Paired rete size is about 2 × 1.5 × 0.2 cm.

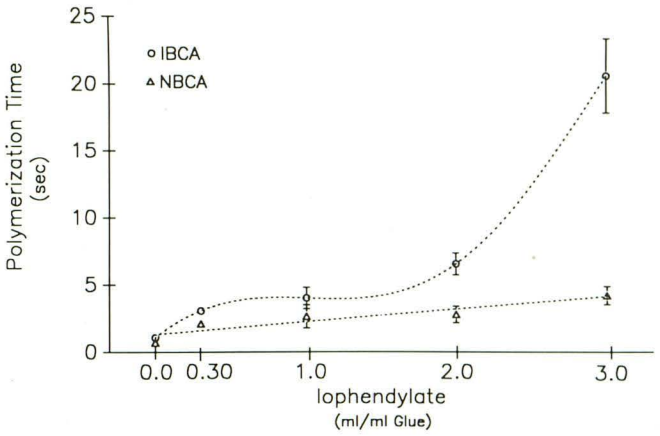


Fig. 2.—Effect of adding iophendylate oil on polymerization time of NBCA and IBCA.

case, to provide a control. Sections were stained with both hematoxylin and eosin and Movat's Pentachrome (for elastin) techniques.

Results

In Vitro Studies

The blue color of Histoacryl became minimal on polymerization, and so did not contribute to visual opacity. Results of the glue alone, glue/oil, and glue/oil/acid polymerization time studies are summarized in Figures 2 and 3. The polymerization time of glue alone is shown at the 0.0 iophendylate position in Figure 2. For each differently proportioned mixture, the means of each glue were statistically significantly different (t test, $p < .001$). Each point on the graph represents a mean of on average 20 observations. Consistently, NBCA showed faster polymerization than IBCA, both in the "pure" state (wherein polymerization times were approximately 0.7 vs 1.0 sec, respectively) and with all subsequent dilutions by oil. Oil had a nonlinear effect on IBCA polymerization time, with marked, nearly exponential prolongation seen when more than a 50/50 ratio was employed. In comparison, only minimal linear prolongation of polymerization time was seen with NBCA using oil dilutions in the clinically relevant range.

Figure 3 summarizes the results of polymerization time studies in which a graded addition of glacial acetic aid to standardized oil/glue mixtures (0.3 ml oil/1 ml glue) was performed. Both glues showed a predictable linear prolongation of polymerization times. Again, NBCA was always faster than IBCA ($p < .001$), and the slope of the plot indicates that a given aliquot of acetic acid produces a slightly smaller effect in NBCA than in IBCA.

Four NBCA/oil/tantalum mixtures, when left to "stand" in tuberculin syringes, all polymerized "spontaneously" at a mean interval of 80 min (range, 45–120 min). NBCA and oil mixed without tantalum did not show any spontaneous polymerization by 3 weeks. Two of three IBCA/oil/tantalum mixtures did polymerize spontaneously, but not before 3–5 days of observation.

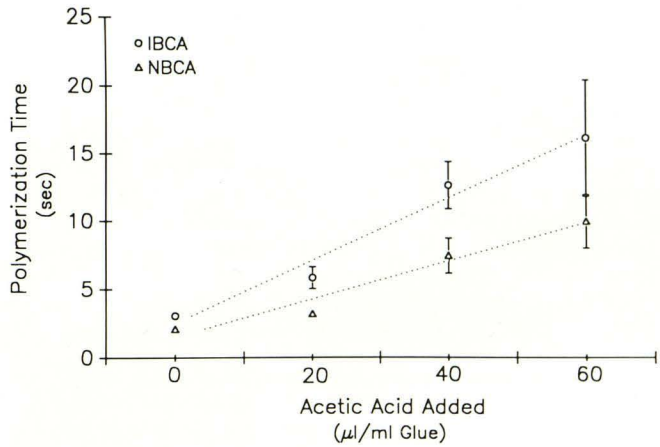


Fig. 3.—Effect of adding glacial acetic acid on polymerization time of NBCA and IBCA. To mimic clinical use, a small amount of iophendylate (0.3 ml/ml glue) was included in all mixtures.

In Vivo Studies: Polymerization Time

While deposition of glue in the ICA was readily achieved in all instances, for subsequent histopathologic study it was essential to fill some of the rete, as this structure's anatomy, consisting of a network of small vessels, provides a model of brain AVM. It was found that a ratio of 0.3 ml oil/1 ml glue was ideal for two reasons: (1) it provided an "effective" polymerization time for both IBCA and NBCA that led to a cast deposition in the distal carotid and at least the proximal portion of the rete when the mixture was trickled into the ICA origin and (2) a low ratio of oil to glue would minimize the oil effect and optimize comparison of the glues' polymerization times.

The site of deposition depended on both the polymerization time and the rate of blood flow carrying the glue. Therefore, it was important to ensure that brisk arterial flow was maintained around the catheter. In 15 of 33 embolized pigs, major brainstem or cerebral infarction occurred, despite a radiographic appearance of incomplete rete filling. It was found empirically that strokes could usually be avoided if the glue did not pass beyond the inferior half of rete (Fig. 1). Animals with any postembolization neurologic deficit were immediately euthanized and therefore excluded from the delayed histologic studies.

Similar results were obtained in all animals studied with approximately equal variations within each glue group (deposition more proximal or more distal than this). No significant difference was demonstrated between the effective polymerization times of the two glues on intraarterial injection for this low oil/glue ratio mixture.

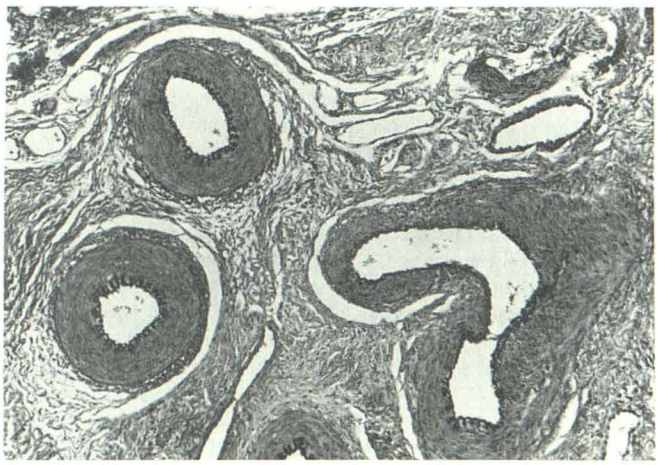


Fig. 4.—Normal rete contralateral to embolus. (Movat, ×122)

In Vivo Studies: Histopathology

The paired rete structure (Fig. 1) measured approximately 2 × 1.5 × 0.2 cm. The embolized side was usually adherent to the surrounding tissues at the skull base, likely due to fibrosis induced by the glue.

Light microscopy of normal rete (Fig. 4) showed well-defined, undulating internal elastic lamina (IEL) and muscular tunica media, with sparse connective tissue between the small vessels. Typical rete vessel lumen diameters ranged from 0.1 to 0.2 mm. Aside from an occasional perivascular inflammatory cell, no pathologic changes were noted in the nonembolized side.

Table 1 summarizes the results of the embolization studies, comparing the histologic reaction at the noted intervals after embolization. Neither glue took up stain, both appearing colorless and slightly refractile. The position of the glue was marked visually by the black tantalum particles, which dispersed along the external margins of the embolus. In animals sacrificed immediately after embolization, the lumen was distended by glue with resultant thinning of the IEL and arterial wall. No inflammatory reaction or vascular injury was evident at this time.

By 2 days postembolization (Fig. 5), a rather intense acute perivascular and/or medial inflammatory reaction, comprising predominantly polymorphonuclear cells, was seen. Frequent foci of intramural hemorrhage were noted in the media, as well as small foci of medial and intimal necrosis. A subintimal dissection was observed with NBCA (Fig. 5B). On days 7 and

TABLE 1: Results of Embolization Studies with IBCA and NBCA in Pigs

Day of Sacrifice	Glue	No. of Pigs	Intensity of Inflammation	Mural Hemorrhage	Mural Necrosis	IEL Defect	Extraluminal Glue	Recanalization
0	IBCA	2	–	–	–	–	–	–
	NBCA	2	–	–	–	–	–	–
2	IBCA	1	+	Recent	Partial	–	–	–
	NBCA	1	+++	Recent[a]	Partial	–	–	–
7	IBCA	1	++	Recent	Partial	Focal	–	–
	NBCA	1	+	Recent	–	Extensive	–	–
14	IBCA	2	++	Recent (1)	Transmural (1)	Extensive (1), focal (1)	Extravascular (1), intramural (1)	–
	NBCA	2	+++	Recent	Partial (1)	Focal (1)	–	–
21	IBCA	1	++	Recent	–	Extensive	–	–
	NBCA	1	+++	Recent	Partial	Focal	Intramural	Yes
28	IBCA	1	+++	Recent and old	Transmural	Extensive	Extravascular	–
	NBCA	1	+	–	–	Focal	Extravascular	–
60	IBCA	3	+++[b]	Old	Transmural (1)	Extensive (2), focal (1)	Extravascular (1)	Yes (1)
	NBCA	3	++[b]	Old (1)	Partial (1)	Extensive (2), focal (1)	Extravascular (2)	Yes (2)

Note.—Intensity of inflammation was graded on a scale of + (mild) to +++ (marked). *Recent* hemorrhage had a visible collection of red cells (hematoma); *old* hemorrhage had hemosiderin/hematoidin pigment. A dash (–) indicates that characteristic was absent. IEL = internal elastic lamina; IBCA = isobutyl 2-cyanoacrylate; NBCA = *n*-butyl 2-cyanoacrylate.

[a] Extensive with fibrosis.

[b] Two pigs had marked fibrosis.

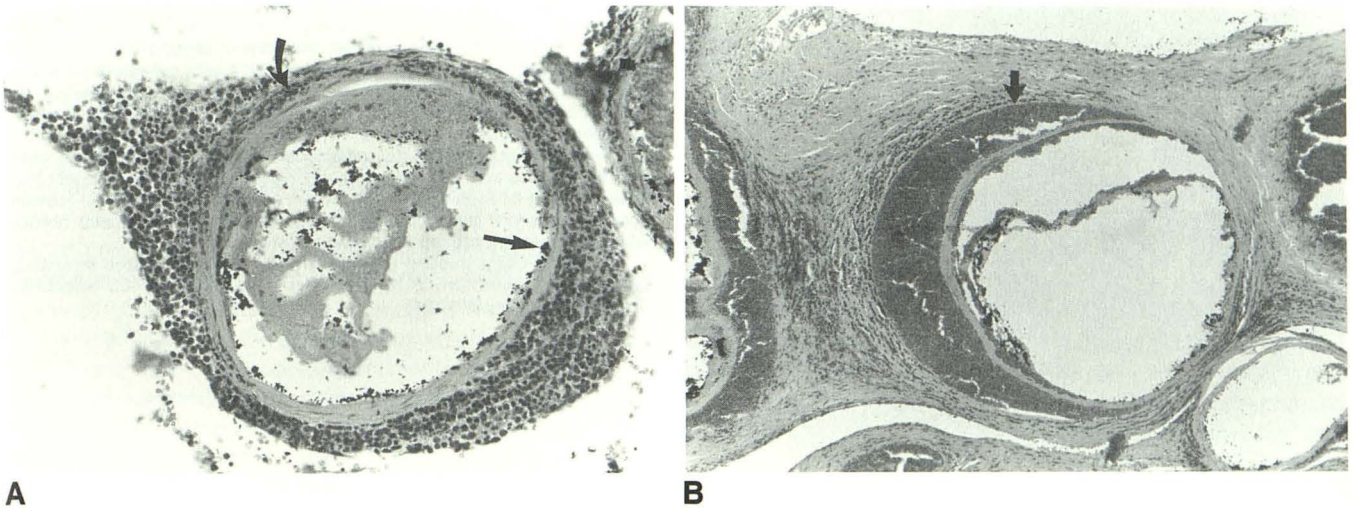


Fig. 5.—2 days after embolization.
***A*, NBCA outlined by black tantalum granules (*straight arrow*). Florid, acute inflammatory infiltrate surrounds this distended vessel and is also present in wall (*curved arrow*). (H and E, ×321)**
***B*, NBCA. Extensive mural hemorrhage dissects layers of embolized vessel wall (*arrow*). (H and E, ×220)**

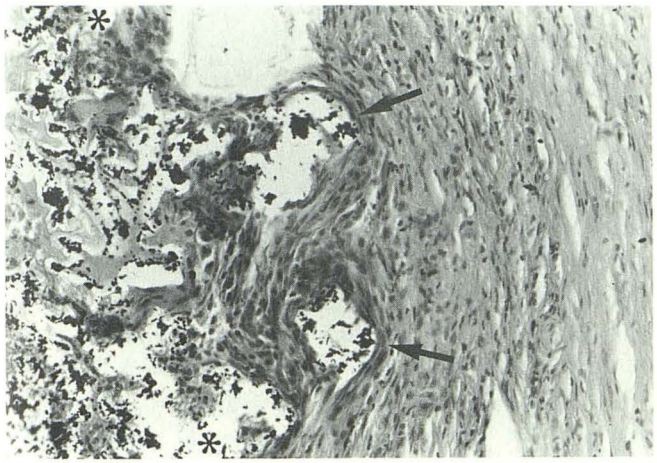


Fig. 6.—2 weeks after embolization. IBCA and tantalum are in two pockets completely sequestered in inner part of vessel wall (*arrows*), surrounded by fibrous tissue. Thrombotic and embolic material lie in lumen on left side of photomicrograph between *asterisks*. (H and E, ×321)

14, similar findings were noted, with frequent focal loss of the IEL in patchy distribution, as well as mural foci of necrosis and hemorrhage. Early foreign-body giant-cell formation was noted around glue emboli at this time. By day 14 the inflammatory reaction intensified, consisting predominantly of transmural round-cell infiltration. The normal architecture of the vessel wall was lost, and the intima destroyed. Portions of the glue became sequestered by fibrous tissue at the glue margins, migrating into the vessel wall (Fig. 6).

On day 21 the inflammation was undiminished and recanalization was first noted. Granulation tissue and fibrosis were variably present, in places quite marked. By day 28, frequent zones of IEL disruption and extensive IEL loss were seen. In some instances, portions of glue were found to be adjacent to these damaged vessels, in an entirely extravascular location. By day 60, a marked foreign-body giant-cell reaction frequently surrounded the intraluminal glue plugs, accompanied by a variable amount of fibrosis. One half of cases at this time revealed glue in a completely extraluminal, extravascular location associated with extensive defects of the IEL. In addition, half the cases showed recanalization of previously occluded lumina; new, multiple, thick-walled vessels were seen coursing past glue plugs (Fig. 7).

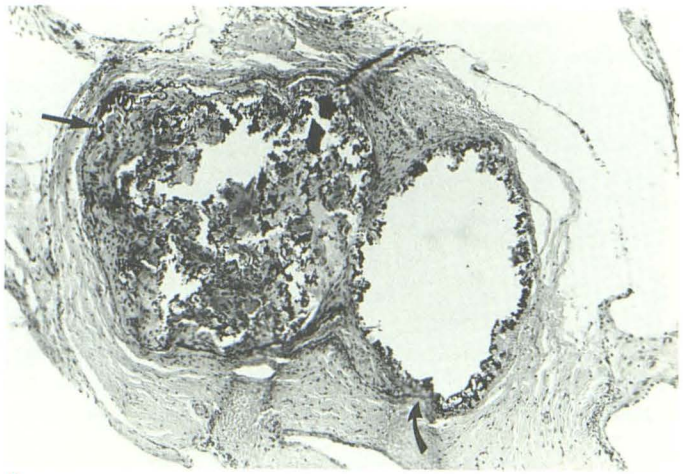
No neoplastic change was seen. The effect of each polymerized glue within the small rete vessels was to provide a "subocclusive thrombus matrix" [20] "entrapping thrombus in its interstices" [23]. The intense vascular injury did not differ between the two glues in type or extent. It can be characterized as an acute and sometimes necrotizing arteritis, becoming a chronic granulomatous arteritis after about 1 month.

Discussion

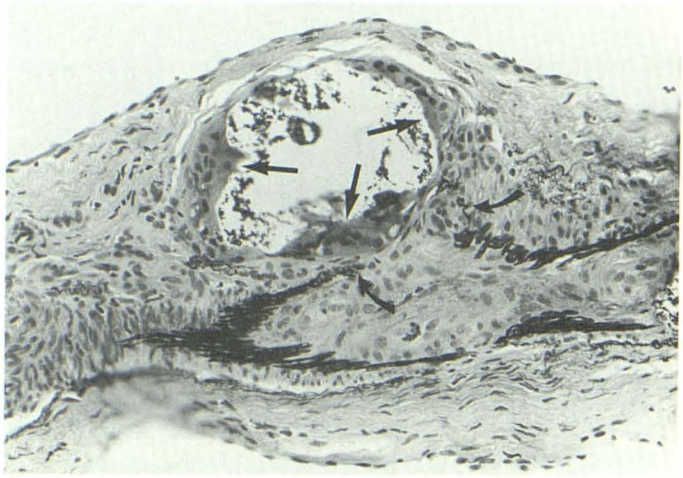
There are few reports of NBCA as an intravascular agent, limited to embolization of abdominal organs and tumors [25–27]. Extensive experience with IBCA embolization has accumulated over the past 10 years [1–9]. Alternative embolic agents are being explored [31–37], but no published study to date has documented efficacy in treatment of brain AVM comparable to that obtained with cyanoacrylic glue.

Polymerization Time

The present study demonstrates significant in vitro differences between NBCA and IBCA that are relevant to their use as embolic agents. Studies of the family of homologues of alkyl-cyanoacrylate [14, 38] have shown an inverse relationship between the alkyl side chain length and the polymeriza-



A



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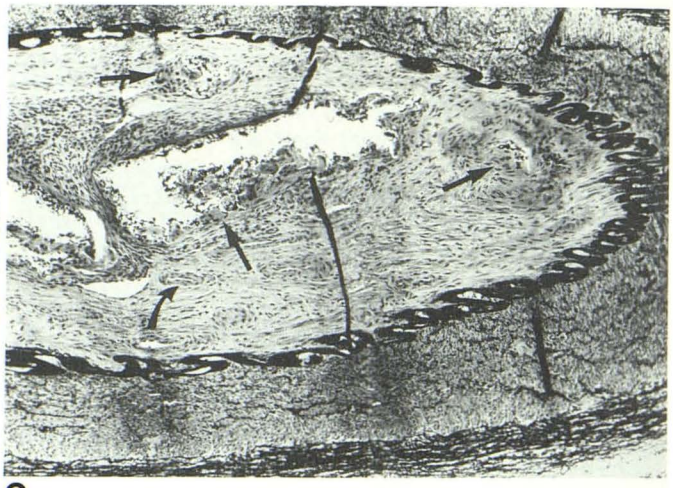

C

Fig. 7.—60 days after embolization.

***A*, IBCA and thrombotic material fill lumen of artery whose internal elastic lamina (IEL) can be traced around most of circumference (*straight arrow*). At right, a large mass of unstained IBCA with peripheral tantalum has extruded through vessel wall, and lies apparently extraluminally. Note broken IEL (*curved arrow*) and glue surrounded by concentric fibrous tissue only. (Movat, ×321)**

***B*, NCBA surrounded by multinucleated foreign-body giant cells (*straight arrows*) and a few strands of fibrous tissue. Embolic agent has been extruded through IEL, whose broken ends are clearly visible (*curved arrows*). Lumen of this obliquely sectioned artery is filled with fibrous tissue representing organized thrombus. (Movat, × 321)**

***C*, NBCA plugs trapped (*straight arrows*) in old organized thrombus, recanalized (*curved arrow*) by numerous small arteries. IEL surrounds former lumen. (Movat, ×321)**

tion time in blood. No direct comparison of NBCA vs IBCA polymerization time has been reported previously.

Good control over the polymerization time of intravascular cyanoacrylate is believed to be of some importance, as the site of glue hardening may determine the efficacy [9], and the morbidity [2], of the embolization procedure. The best long-term results of AVM occlusion can be expected if polymerization occurs within the AVM nidus [39], rather than too proximal in feeding arteries or too distal in draining veins or beyond; also, venous occlusion likely is a cause of hemorrhagic and ischemic complications. Thus, when the delivery microcatheter has been positioned as well as possible in a feeding pedicle, a judgment must be made, based on clinical experience, of where liquid glue injected intravascularly will likely harden. This will depend on the speed of blood flow at that injection site, the length of the pedicle between catheter tip and nidus, and the polymerization characteristics of the glue. Of these three factors, only the latter is readily adjustable in the usual clinical setting, through use of oil or acetic acid added to the glue/tantalum mixtures. (The use of leak balloons to provide flow arrest, and the rate of injection, are additional factors influencing deposition site.)

The minimal effect of iophendylate oil on the polymerization time of NBCA indicates that this method of polymerization time adjustment, so widely used with IBCA, will not be effective. This difference was also suggested indirectly by Stosslein et al. [27], who found that a great excess of added oil was needed to prolong the polymerization time of Histoacryl. Early experience with Histoacryl/oil mixtures in unpublished clinical use in cerebral AVM further supports this finding.

In comparison, prolongation of polymerization time by addition of acetic acid is more predictable and better controlled. The addition of oil to the embolic mixture is still desirable, primarily to aid in the suspension of the tantalum, but no more than 0.3 ml/ml glue is necessary. Control of polymerization time by addition of acetic acid appears more consistent than was possible with oil [20, 21], with lower viscosity and ease of injection as additional benefits [9, 29].

The absolute values of polymerization times found in our study in static plasma are not directly applicable to clinical use, as the mixing of glue molecules with anions, which likely occurs on intravascular injection [29], results in more rapid polymerization. Also, polymerization of cyanoacrylates at body temperature is faster than at room temperature [40],

acting to further accelerate the times of both glues. As can be seen from inspection of Figures 2 and 3, as polymerization time decreases, the differences between the two glues is progressively minimized. These mechanisms presumably explain the discrepancy between the in vivo and in vitro results: the speed advantage of NBCA polymerization in static room temperature plasma is lost on intraarterial injection, because polymerization of both glues is accelerated. When polymerization prolongation is not desired, the clinical behavior of the two glues should be similar on intravascular injection, if such a low oil/glue ratio (0.3 ml/1.0 ml) mixture is used.

The mechanism of the spontaneous polymerization of both NBCA and IBCA after contact with tantalum is unclear: this effect has not been previously described. The initiation of polymerization of cyanoacrylates can occur by either anionic or free-radical mechanisms, the essential step being addition of a negative ion to open the carbon-carbon double bond of the monomer [40]. The polymer is then propagated through the resultant negatively charged species. This is the basis for polymerization inhibition by acetic or other added acids, which reduce transiently the local availability of anions; acids may also terminate a propagating polymerization chain, until they are consumed in the process [29].

Known polymerization initiators include any source of a hydroxyl ion, such as water or methanol; X-rays and ultraviolet light; as well as biological amines and amino acids. These reactions all proceed quickly, whereas we observed a very long induction period in this "tantalum effect," particularly in IBCA. Presumably, the tantalum is capable of donating electrons to initiate the reaction, acting as a weak catalyst. This phenomenon has clinical relevance: mixtures of NBCA with tantalum should be used or discarded within 15 min of preparation to avoid problems. It is unlikely that tantalum speeds polymerization of the mixture when injected intraarterially within this time. If this were the case, the speed advantage of NBCA would have been further exaggerated, and likely produced a demonstrably fast polymerization time in our carotid/rete study.

Why iophendylate or other oils should slow cyanoacrylate polymerization has not been well explained. A possible mechanism is reduced anion availability due to the glue being "dissolved" in oil, thereby transiently preventing access to blood ions until sufficient mixing has occurred to "expose" the monomer [29]. The physicochemical characteristics of the various alkyl homologues of cyanoacrylate differ in some respects; whether, for example, the degree of lipid solubility might influence the magnitude of the oil effect is unknown.

Essentially all available prepared cyanoacrylate adhesives contain additives, including stabilizers and/or acidic inhibitors [40], that may affect polymerization time. The degree to which our results might differ from those obtained by using pure monomers of NBCA and IBCA is uncertain, but has little relevance, since pure monomers are not clinically useful on account of their instability.

Histopathology

In order to consider NBCA as a potential agent for human embolization, two major issues must be addressed. First, it must be shown that the histopathologic reaction in tissue embolized with NBCA is less than, or at worst no more severe than, that seen with IBCA. Secondly, the question of potential carcinogenesis raised with IBCA is no less important for NBCA, as the two are chemically similar.

No previous study has directly compared the histotoxicity of IBCA and NBCA in either surgical or intravascular use. The histopathology of IBCA emboli in various animal and human tissues has recently been reviewed [8]. Although the tissue reactions induced by surgical application of NBCA to wound edges have been described [13–15], those of NBCA emboli have not, aside from mention of foreign-body giant-cell reactions around the embolus [25, 26]. For IBCA emboli, typical appearances described include an initial acute, transmural, but variable degree of focal inflammation, progressing to a loss of wall layer definition and disruption of the IEL. Gradually, lymphocytes replace polymorphs, and later foreign-body giant cells and fibrosis predominate [8]. The intensity of the vascular injury in animal studies has been particularly inconsistent. The best-documented changes are those described by Vinters et al. [21] in human cerebral AVM specimens previously embolized with IBCA. Their findings in part coincide with those of two animal studies that describe a severe vascular injury induced by IBCA emboli, in dog renal artery [18] and pig visceral arteries [22].

We have identified six phenomena of particular interest in our specimens: inflammation, intramural hemorrhage, mural necrosis, breaks in the IEL, extravasation of glue, and recanalization (Table 1). We found no significant difference between NBCA and IBCA in any of these features.

The pattern and time course of inflammation and subsequent fibrosis that we have observed coincides with that in other studies [18, 21, 23]. The degree of variability in intensity of histopathologic reaction to each glue was striking. This may explain the inconsistency of reported results of previous animal studies that used only a few animals each. Foci of mural hemorrhage have not been previously reported in either animal studies or embolized human AVMs. Significantly, relatively fresh foci of mural hemorrhage were seen as late as 1 month, and evidence of old and recent hemorrhage could be found simultaneously. A continuing acute process is suggested by these findings. Patchy, focal mural necrosis has been noted by other authors, as have the breaks in the IEL [18, 21, 22].

The rete may be uniquely sensitive to vascular injury by these embolic materials. It differs from other normal vascular beds in that, like an AVM, its inflow and outflow blood pressures are at arterial levels [30]. Routinely, embolization of the rete occludes only a portion of the vascular network while multiple collaterals from external carotid and other branches at the skull base [30] maintain flow in unembolized portions. Thus, there is no territory of infarction on occlusion; instead there is a continuing demand distally for flow, likely acting to promote recanalization. Whatever the reason for the intensity of the histopathologic reaction to glue in pig rete, the model seems ideally suited to demonstrate any slight difference that might exist between two different glues; yet no such difference was found.

The two most striking findings were the sequestration and eventual extrusion of glue into the extravascular space and the recanalization of lumina previously occluded by glue. Both these features have been described in human brain AVM after embolization [21], but not in previous animal studies or in other human embolized vessels. Overall, the pig rete and human cerebral AVM appear to respond in a virtually identical fashion to cyanoacrylate injury.

These phenomena have further clinical importance. It is widely held that cyanoacrylic glue is the most permanent of the embolic agents. Yet, clinical experience with incompletely embolized brain AVMs has shown that regrowth or recanalization of the AVM is common [9]; while this may be partly due to recruitment of new feeders, the histologic findings in our study, together with those of Vinters et al. [21], suggest another possible explanation: that occlusion of a vascular lumen by glue is not permanent. In our specimens and in those of other authors [18, 19, 21, 23], polymerized glue only partially fills the injected lumen. It has been estimated that 15% or less of the glue embolus is in contact with the endothelial surface [23]. The remainder of the lumen becomes occluded by thrombus (a process induced vigorously by glue), and subsequently organized. It is clear, however, that such an occlusion does not remain stable. Instead, portions of glue become sequestered from the main mass, and migrate through the vessel wall, likely via breaks in the IEL. As well, angiogenesis and recanalization occur within the occluded lumen, even immediately adjacent to the glue embolus.

Despite disruption of the vessel wall layers and glue extrusion, no case of local hemorrhage from the embolized rete vessels was seen. Vinters et al. [8] have suggested that such a mechanism could account for delayed hemorrhage after embolization of human cerebral AVM with glue. The normal rete vessels are relatively thick-walled compared with the abnormal vascular channels in AVM; yet, after chronic embolization, similar degrees of transmural inflammation, damage, and necrosis were found. The intense fibrotic response associated with glue plugs in some specimens would seem to offer a degree of protection from hemorrhage. It is of some interest that the presence of glue extrusion did not correlate well with active intense inflammation (Table 1).

The mechanism of tissue toxicity has been debated [8]. It is unlikely to be due to the heat produced in the exothermic polymerization reaction, as this amounts to no more than a few degrees locally [17, 41]. An ischemic cause is unlikely, as we have observed no vasovasorum in rete walls. Toxic degradation products may be responsible. Methyl cyanoacrylate has been reported to break down in vitro into formaldehyde and cyanoacetate, both toxic to tissues [41, 42]. In vivo, poly-IBCA degrades to isobutyl alcohol and polycyanoacrylic acid [43].

Various animal studies have shown that cyanoacrylates persist in tissues for months to years, the duration proportional to the length of the alkyl side chain [17, 40, 44]. This property may explain the reduced local toxicity of the longer alkyl homologues, as toxic by-products are formed more slowly and therefore better tolerated [17]. However, it is this quality of tissue persistence that has given rise to concerns of possible carcinogenicity. No human case of carcinogenesis has occurred in over 15 years of clinical cyanoacrylic use [10]. Animal studies have revealed conflicting findings, but none have used intravascular glue injection. Large doses of NBCA induced tumors in rats in one study [45], but in others, neither IBCA nor NBCA did so [15, 46, 47]. Carcinogenesis is unlikely to occur with neurointerventional cyanoacrylate use, for several reasons. Typical total glue volumes used in the treatment of human AVM do not exceed 1–2 ml (and are often less). Proportionately, such a dose is miniscule on the basis of body weight, when compared with that used in rat studies [10]. As well, the site of deposition can significantly alter the risk of carcinogenesis, for example, epidural and subdural implants produced no cancers, while in the same rat species, subcutaneous ones did [48].

It could be argued that even a remote possibility of carcinogenesis should preclude the use of an embolic agent, and on this basis some might instead choose a particulate agent, such as polyvinyl alcohol (Ivalon). Polyvinyl alcohol is now probably the most frequently used alternative to cyanoacrylate for embolization of AVMs [7, 32–34, 36, 49–53]. Yet, it, too, has been shown to be carcinogenic in animal studies [54–56]. Thus, the issue of theoretical risk in human use will not be eliminated by substituting polyvinyl alcohol for adhesives.

We use very small doses of NBCA clinically (typically 0.2–0.4 ml per patient), usually as an adjunct to embolization with particulate agents, mainly in cases of fistulous arteriovenous flow [57]. Brain AVM embolization holds other more definite risks for the patient [2, 3, 7] that are discussed and weighed before the decision to treat is made. That a remote, theoretical risk of carcinogenesis be included in an informed consent is difficult to justify, unless that type of risk be included in an informed consent for virtually every other medical act where theoretical carcinogenicity has not been absolutely excluded.

We believe that the continued use of NBCA is mandated when alternative embolic agents are ineffective and embolization is clinically indicated.

Conclusions

The inflammatory reactions induced by IBCA and NBCA emboli are indistinguishable. They are typified by a vasculitis that is initially acute and sometimes necrotizing. Within a month or so, a chronic granulomatous vasculitis supervenes. The severity and extent of reaction is quite variable. The IEL is frequently disrupted or lost extensively. This is associated with migration of portions of glue into the vessel wall and perivascular space. Furthermore, recanalization occurs around glue plugs. Thus, it appears that vascular occlusion with cyanoacrylate glue is not necessarily permanent.

In summary, NBCA, while causing significant vascular injury, is not different from IBCA in this respect. It is, however, demonstrably more reactive: it is polymerized more easily and quickly than IBCA and the polymerization speed is affected much less by the addition of oil. When the need for such fast-polymerizing embolic agents is compelling, NBCA could be directly substituted for IBCA, requiring little adjustment in clinical technique: (1) NBCA mixtures should be used within 15 min of preparation, as contact with tantalum can induce

polymerization as early as 45 min; (2) the addition of iophendylate oil, in proportions commonly used with IBCA, will not effectively prolong polymerization time; (3) oil addition is still useful to suspend the tantalum; (4) low oil/glue ratio mixtures (0.3 ml/1.0 ml) of NBCA or IBCA polymerize at a similar rate on intravascular injection, permitting direct substitution of NBCA for IBCA in clinical use; and (5) polymerization time is readily adjusted by glacial acetic acid, and the effect is more predictable in either glue than was possible by oil alone in IBCA.

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REFERENCES

1. Cromwell LD, Harris AB. Treatment of cerebral arteriovenous malformations: a combined neurosurgical and neuroradiological approach. *J Neurosurg* **1980**;52:705–708
2. Debrun G, Viñuela F, Fox AJ. Embolization of cerebral arteriovenous malformations with bucrylate. Experience in 46 cases. *J Neurosurg* **1982**; 56:615–627
3. Pelz D, Fox AJ, Viñuela F, Drake CC, Ferguson GG. Preoperative embolization of brain AVMs with isobutyl-2 cyanoacrylate. *AJNR* **1988**;9: 757–764
4. Spetzler RF, Martin NA, Carter LP, Flom RA, Raudzens PA, Wilkinson E. Surgical management of large AVM's by staged embolization and operative excision. *J Neurosurg* **1987**;67:17–28
5. Viñuela F, Fox AJ. Interventional neuroradiology and the management of arteriovenous malformations and fistulas. *Neurol Clin* **1983**;1(1):131–154
6. Luessehop AJ, Rosa L. Cerebral arteriovenous malformations: indications for and results of surgery and the role of intravascular techniques. *J Neurosurg* **1984**;60:14–22
7. Dubois PJ, Kerber CW, Heinz ER. Interventional techniques in neuroradiology. *Radiol Clin North Am* **1979**;17(3):515–542
8. Vinters HV, Galil KA, Lundie MJ, Kaufmann JCE. The histotoxicity of cyanoacrylates. *Neuroradiology* **1985**;27:279–291
9. Viñuela F, Fox AJ, Pelz D, Debrun G. Angiographic follow-up of large cerebral AVMs incompletely embolized with isobutyl-2-cyanoacrylate. *AJNR* **1986**;7:919–925
10. Berenstein A, Hieshima G. Clinical versus experimental use of isobutyl-2-cyanoacrylate (letter). *J Neurosurg* **1987**;67:318–319
11. Samson D, Marshall D. Carcinogenic potential of isobutyl-2-cyanoacrylate (letter). *J Neurosurg* **1986**;65:571–572
12. Samson D, Marshall D. The use of isobutyl-2-cyanoacrylate in embolization (letter). *Surg Neurol* **1987**;28:319
13. Gyurko G, Szucs J, Ezsely F, Koja C, Bednarsky Z. Use in vascular surgery of the tissue adhesive Histoacryl. *Acta Chir Hung* **1974**;15(4):353–360
14. Matsumoto T, Hardaway RM III, Heisterkamp CA III, Vani RC, Leonard F. Higher homologous cyanoacrylate tissue adhesives in surgery of internal organs. *Arch Surg* **1967**;94:861–864
15. Matsumoto T, Nemhauser GM, Soloway HB, Heisterkamp C, Auby G. Cyanoacrylate tissue adhesives: an experimental and clinical evaluation. *Milit Med* **1986**;134:247–252
16. Lehman RAW, Hayes GJ. The toxicity of alkyl-2-cyanocrylate tissue adhesives: brain and blood vessels. *Surgery* **1967**;61(6):915–922
17. Woodward SC, Herrman JB, Cameron JL, Brandes G, Pulaski EJ, Leonard F. Histoxicity of cyanoacrylate tissue adhesive in the rat. *Ann Surg* **1965**;162(1):113–122
18. Cromwell LD, Freeny PC, Kerber CW, Lunz LL, Harris AB, Shaw CM. Histologic analysis of tissue response to bucrylate-Pantopaque mixture. *AJR* **1986**;147:627–631
19. Freeny PC, Mennemeyer R, Kidd CR, Bush WH. Long-term radiographic-pathologic follow-up of patients treated with visceral transcatheter occlusion using isobutyl 2-cyanoacrylate (bucrylate). *Radiology* **1979**;132: 51–60
20. Kish KK, Rapp SM, Wilner HI, Wolfe D, Thomas LM, Barr J. Histopathologic effects of transarterial bucrylate occlusion of intracerebral arteries in mongrel dogs. *AJNR* **1983**;4:385–387
21. Vinters HV, Lundie MJ, Kaufmann JCE. Long term pathological followup of cerebral arteriovenous malformations treated by embolization with bucrylate. *N Engl J Med* **1986**;314:477–483
22. White RI, Strandberg JV, Gross GS, Barth KH. Therapeutic embolization with long term occluding agents and their effects on embolized tissues. *Radiology* **1977**;125:677–687
23. Zanetti PHY, Sherman FE. Experimental evaluation of a tissue adhesive as an agent for treatment of aneurysms and arteriovenous anomalies. *J Neurosurg* **1972**;36:72–79
24. Katada K, Saro H, Katoh Y, et al. Ethyl 2-cyanoacrylate as an embolic agent for cranial arteriovenous malformations. *Acta Radiol [Suppl]* **1986**; 369: 623–626
25. Gunther R, Schubert U, Bohl J, Georgi M, Marberger M. Transcatheter embolization of the kidney with butyl-2-cyanoacrylate: experimental and clinical results. *Cardiovasc Intervent Radiol* **1987**;1:101–108
26. Rossweiler J, Kauffman GW, Richter G, Fuchs G, Miller K. Experimental basis of angioinfarction for the treatment of renal hypertension. *Urol Int* **1986**;41:42–56
27. Stosslein F, Ditscherlein G, Romaniuk PA. Experimental studies on new liquid embolization mixtures (Histoacryl-Lipiodol, Histoacryl-Pantopaque). *Cardiovasc Intervent Radiol* **1982**;5:264–267
28. Cromwell LD, Kerber CW. Modification of cyanoacrylate for therapeutic embolization: preliminary experience. *AJR* **1979**;32:799–801
29. Spiegel SM, Viñuela F, Goldwasser JM, Fox AJ, Pelz DM. Adjusting the polymerization time of isobutyl-2 cyanoacrylate. *AJNR* **1986**;7:109–112
30. Du Boulay G. Comparative neuroradiologic anatomy of experimental animals. In: Newton TH, Potts DG, eds. *Radiology of the skull and brain*. St. Louis: Mosby, **1974**:2782–2783
31. Berenstein A, Russell E. Gelatin sponge in therapeutic neuroradiology: a subject review. *Radiology* **1981**;141:105–112
32. Dion J, Vinuela F, Lylyk P, Lufkin P, Bentson J. Ivalon-33% ethanol-Avitene embolic mixture: clinical experience with neuroradiological endovascular therapy in 44 arteriovenous malformations (abstr). *AJNR* **1988**;9:1029
33. Fox AJ, Lee DH, Pelz DM, Brothers MF, Deveikis JP. A thrombotic mixture as a "polymerizing" agent (abstr). *ANJR* **1988**;9:1029
34. Horton JA, Marano GD, Kerber CW, Jenkins JJ, Davis S. Polyvinyl alcohol foam–Gelfoam for therapeutic embolization: a synergistic mixture. *AJNR* **1983**;4:143–147
35. Lee DH, Wriedt CR, Lylyk P, Fox AJ, Vinuela F, Kaufmann JCE. Thrombotic embolization agents: tests in pig rete (abstr). *AJNR* **1987**;8:937
36. Smith HJ, Strother CM, Kikuchi Y, et al. MR imaging in the management of supratentorial intracranial AVMs. *AJNR* **1988**;9:225–235
37. Strother CM, Laravusco R, Rappe A, Su SL, Northern K. Glutaraldehyde cross-linked collagen (GAX): a new material for therapeutic embolization. *AJNR* **1987**;8:509–515
38. Collins JA, Pani KC, Lehman RA, Leonard F. Biologic substrates and cure rates of cyanoacrylate tissue adhesives. *Arch Surg* **1966**;93:428–432
39. Drake CG. Cerebral arteriovenous malformations: indications for and experience with surgical treatment in 166 cases. *Clin Neurosurg* **1979**; 26:145–208
40. Matsumoto T. The chemistry of cyanoacrylate adhesives. In: Matsumoto T, ed. *Tissue adhesives in surgery*. Flushing, NY: Medical Examination Publishing Co., **1972**:154–188
41. Leonard F. Physical and chemical aspects of cyanoacrylate tissue adhesives. In: *Adhesives in surgery. Proceedings of the international symposium.* Vienna: Wiener, 1968:11–19
42. Leonard F, Kulkarni RK, Brandes GI, Nelson J, Cameron JJ. Synthesis and degradation of poly (alkyl-2-cyanoacrylates). *J Appl Poly Sci* **1966**;10: 259–272
43. Lenaerts V, Couvreur P, Christiaens-Leyh D, et al. Degradation of poly (isobutylcyanoacrylate) nanoparticles. *Biomaterials* **1984**;5:65–68
44. Pani KC, Gladieux G, Brandes G, Kulkarni RK, Leonard F. The degradation of N-butyl-alpha cyanoacrylate tissue adhesive. *Surgery* **1968**;63:481–489
45. Reiter A. Sarkomerzeugen de Wirkung des Gewebeklebers Histoacryl-blau an der Ratte. *Z Exp Chir Transplant Kunstliche Organe* **1987**;20(1): 55–60
46. Matsumoto T, Heisterkamp CA. Long-term study of aerosol cyanoacrylate tissue adhesive spray: carcinogenicity and other untoward effects. *Am*

Surg **1969**;35(11):825–827
47. Heiss WH. The use of synthetic polymeric materials as suture substitutes and their place in pediatric surgery. *Prog Pediatr Surg* **1979**;1:99–150
48. Kamijyo Y. EDH adhesive: tissue reaction and carcinogenicity of EDH-adhesive and other plastics in animals. *Nippon Geka Hokan* **1968**;37: 204–230
49. Jack CR, Forbes G, Dewanjee MK, Brown ML, Earnest F IV. Polyvinyl alcohol sponge for embolotherapy: particle size and morphology. *AJNR* **1985**;6:595–597
50. Lanman TH, Martin NA, Vinters HV. The pathology of encephalic arteriovenous malformations treated by prior embolotherapy. *Neuroradiology* **1988**;30:1–10
51. Latchaw RE, Gold LHA. Polyvinyl foam embolization of vascular and neoplastic lesions of the head, neck and spine. *Radiology* **1979**;131: 669–679
52. Spetzler RF, Martin NA, Carter LP, Flom RA, Raudzens PA, Wilkinson E. Surgical management of large AVM's by staged embolization and operative excision. *J Neurosurg* **1987**;67:17–28
53. Todavarthy SM, Moller JH, Amplatz K. Polyvinyl alcohol (Ivalon)—a new embolic material. *AJR* **1975**;125(3):609–616
54. Hueper WC. Carcinogenic studies on water-soluble and insoluble macromolecules. *Arch Pathol Lab Med.* **1959**;67:589–617
55. Jukes CE, Mitchley BCV. Polyvinyl sponge implants: experimental and clinical observations. *Br J Plast Surg* **1962**;15:225–235
56. Oppenheimer BS, Oppenheimer ET, Danishefsky I, Stout AP, Eirich FR. Further studies of polymers as carcinogenic agents in animals. *Cancer Res* **1955**;15:333–340
57. Vinuela F, Fox AJ, Pelz DM. Identification of arteriovenous fistulae in cerebral AVMs: important therapeutic implications (abstr). *AJNR* **1986**; 7:553