Hemodialysis patient survival is dependent on the availability of a reliable vascular access. In clinical practice, procedures for vascular access cannulation vary from clinic to clinic. We investigated the impact of cannulation technique on arteriovenous fistula and graft survival. Based on an April 2009 cross-sectional survey of vascular access cannulation practices in 171 dialysis units, a cohort of patients with corresponding vascular access survival information was selected for follow-up ending March 2012. Of the 10,807 patients enrolled in the original survey, access survival data were available for 7058 patients from nine countries. Of these, 90.6% had an arteriovenous fistula and 9.4% arteriovenous graft. Access needling was by area technique for 65.8%, rope-ladder for 28.2%, and buttonhole for 6%. The most common direction of puncture was antegrade with bevel up (43.1%). A Cox regression model was applied, adjusted for within-country effects, and defining as events the need for creation of a new vascular access. Area cannulation was associated with a significantly higher risk of access failure than rope-ladder or buttonhole. Retrograde direction of the arterial needle with bevel down was also associated with an increased failure risk. Patient application of pressure during cannulation appeared more favorable for vascular access longevity than not applying pressure or using a tourniquet. The higher risk of failure associated with venous pressures under 100 or over 150 mm Hg should open a discussion on limits currently considered acceptable.
needles and low blood flow rates of 200–250 ml/min are generally recommended, particularly in AVFs. In mature accesses, larger 15- or 14-G needles are required to support the higher blood flow rates of ≥350 ml/min needed for high-efficiency dialysis or convective treatments. There is some concern that high blood flow may have a negative impact on access survival. Thus, the influence of needle size on access patency remains an open subject.

Various other aspects of cannulation are not addressed in the guidelines, and there exist a variety of options to choose from. For example, standard double-needle cannulation involves inserting two large bore needles into the fistula or graft vessels, whereby three distinctly different methods for puncture site selection exist: area, rope-ladder, and buttonhole. Area cannulation refers to puncturing of the same general area session after session. In the rope-ladder technique, the cannulator changes the needle placement sites for each dialysis, choosing sites at a defined distance along the VA line from the previous puncture sites. In the buttonhole method of cannulation, hemodialysis needles are inserted in the exact same spot and at the same angle and depth of penetration for consecutive dialyses. The venous needle returns the blood from the extracorporeal circuit to the body and must always point in the direction of blood flow (antegrade direction). The arterial needle is used to withdraw blood from the vasculature to the extracorporeal circuit and may point either in the same direction of blood flow (i.e., antegrade direction) or in the opposite direction (i.e., retrograde direction) (Figure 2). The optimal direction of arterial access needles in fistulas and grafts remains a subject of some controversy.14,15

Whether or not to exert arm pressure at the time of cannulation, either using a tourniquet or manual pressure, is a further subject of debate, although application of a tourniquet is recommended by KDOQI.13

Despite previous studies having addressed the issue of VA techniques and associated access survival, to date, there is a lack of convincing evidence supporting one particular cannulation procedure or a combination of procedures.16,17 The primary aim of this study is to investigate whether diverse aspects of AVF and arteriovenous graft cannulation have an effect on access longevity and are consequentially more or less recommendable. Furthermore, as blood flow, venous pressure, and location of the access (e.g., right or left arm, distal or proximal) have been discussed in association with access patency, the relationship between these and access survival is also addressed as a secondary aim.18

RESULTS
Out of the 10,807 patients enrolled for the original survey, access survival data were available for 7058 (65%) patients. These patients resided in Portugal, the United Kingdom, Ireland, Italy, Turkey, Romania, Slovenia, Poland, and Spain. The mean age was 63.5 ± 15.0 years, 38.5% were female, 27.1% were diabetic, 90.6% had a native fistula, and 9.4% had a graft. Median dialysis vintage was 43.2 months (minimum: 0.1 months; maximum: 419.6 months). Access location was lower arm for 51.2% of patients. During the follow-up, 51.1% were treated with antiaggregants and 2.8% with anticoagulants. Local anesthesia was commonly exercised in the United Kingdom and Ireland only. Prevalent needle sizes were 15 and 16 G for 63.7% and 32.2% of the patients, respectively (14 G: 2.7%; 17 G: 1.4%). In Spain, 98% of patients were treated with 15-G needles, and in Romania 75% of patients were treated with 16-G needles. Cannulation technique was area for 65.8%, rope-ladder for 28.2%, and buttonhole for 6% of patients, with some country preferences clearly visible: area technique was applied in as much as 77% of patients in Romania, and rope-ladder was more common in Poland than in the total study population (44%). The direction of arterial puncture was antegrade for 57.3% of patients; this was the preference for 99% of patients in Poland. The bevel orientation was upward for 70.2% of the patients, peaking in Poland with 95%. The practice of needle rotation after insertion was practiced for 42% of patients, with a much higher percentage in Italy (82%). The prevalent
Combination between arterial needle puncturing and bevel direction was antegrade with bevel upward (43.1%), followed by retrograde with bevel up (27.1%). The proportion of the two other combinations, that is, antegrade and retrograde with bevel downward, was 14.2% and 15.6%, respectively. The 15.6% with retrograde and bevel down were mainly treated in two countries (Spain and Portugal). Median blood flow was 350–400 ml/min. In Italy and Spain, 40% and 38% of patients were treated with blood flows exceeding 400 ml/min, respectively. Conversely, in Slovenia and Poland 54–55% of patients were treated with blood flows below 300 ml/min. Figure 3 shows the distribution of patients according to the prescribed needle size, blood flow, and venous pressure levels. An association between needle size, blood flow, and venous pressure is transparent.

The primary outcome event (i.e., surgery for a new VA during the follow-up period) was observed in 1485 patients (21%). The majority of these events were due to thrombosis. Univariate survival analysis revealed a significant benefit for access survival for patients who are younger, nondiabetic, male, have lower body mass index, do not take platelet antiaggregants, do not have heart failure, and are able to assist with compression. A significant benefit was also seen for patients with fistula (vs. graft), smaller needles, distal location of the access, and low venous pressure. With regard to cannulation technique, positive effects were observed for antegrade needle direction (vs. retrograde), bevel up (vs. down), nonalcohol-based disinfection, and application of local anesthesia. Although not statistically significant, a potential survival benefit was indicated for higher blood flow ($P = 0.056$) and buttonhole technique (vs. rope-ladder and area, $P = 0.11$). Needle rotation did not affect the access survival ($P = 0.81$), neither did access vintage (age $< 1$ month before baseline vs. $\geq 1$ month before baseline; $P = 0.29$). The Kaplan–Meier access survival curves according to blood flows, venous pressures, needle sizes, and cannulation techniques are presented in Figure 4.

In a second step, a multivariable Cox regression model was calculated (Table 1). After adjustment for age, gender, diabetes, VA type, access location (proximal vs. distal), dialysis vintage and heart failure, and incorporation of country differences, the use of a 16-G needle was associated with a significantly higher risk of access failure (hazard ratio (HR) 1.21) compared with the use of a 15-G needle. Very few (1.4%) patients were treated with the even smaller 17-G needles, but the direction of the results is the same, that is, increased HR for smaller needle size. Using a blood flow of 300–350 ml/min as a reference, the HR tended to decrease as the blood flow increased. With regard to cannulation technique, both rope-ladder and buttonhole techniques performed significantly better than the area technique. Considering antegrade with the bevel up as reference, the retrograde direction of the arterial needle with bevel down is associated with a significant increase of access failure risk of 18%. All other options, that is, antegrade direction with bevel down or retrograde direction with bevel up, were not associated with a HR significantly different from 1.00. With regard to venous pressure, using as reference the range between 100 and 150 mm Hg, the HRs increased proportionally to 1.4, 1.87, and 2.09 with the increase of venous pressure from 150 to 200 mm Hg, 200 to 300 mm Hg, and $> 300$ mm Hg, respectively (all $P \leq 0.008$). Of note, venous pressures of

![Figure 3](image-url)
>300 mm Hg are extreme cases and were only recorded in 0.6% of the patients. In addition, a venous pressure of <100 mm Hg was associated with a significantly higher HR of 1.51. To investigate this further, we also looked for interaction effects between blood flow and venous pressure, as well as between arterial and venous pressures; no significant associations were found.

Finally, the use of a tourniquet and not applying any pressure at the time of cannulation were associated with HRs of 1.30 and 1.25 ($P<0.008$ and $<0.02$), respectively, compared with exertion of arm compression by the patient at the time of cannulation (labeled ‘patient assistance’ in Table 1).

**DISCUSSION**

In summary, this study revealed that area cannulation technique, albeit being identified as the most commonly used technique in this population of over 7000 patients, was inferior to rope-ladder and to buttonhole for maintenance of VA functionality. With regard to the effect of needle and bevel direction, the combination of antegrade positioning of the arterial needle with bevel-up orientation was significantly associated with better access survival than retrograde positioning with bevel down. The use of larger needles tended to favor access patency, with 15 G being superior to 16 or 17 G. The application of arm pressure by the patient at the time of cannulation had a favorable effect on access longevity compared with not applying pressure or using a tourniquet. Results pertaining to the type and location of the access and the technical parameters (i.e., blood flow and venous pressure) were as follows: there was an increased risk for access failure for grafts vs. fistulas, proximal location vs. distal, right arm vs. left arm, blood flows below 300 ml/min vs. those in the range of 300–350 ml/min, and for the presence of a venous pressure >150 mm Hg vs. pressures between 100 and 150 mm Hg. Tissue reparative processes triggered by cannulation procedures may cause enlargement of the fistula and the formation of aneurisms and scars that, in turn, can favor the development of stenotic lesions and ultimately impact fistula survival. Repetitive punctures at the VA site cause vessel wall defects that are initially filled by thrombi before finally healing. Of the three cannulation techniques, the buttonhole approach has the theoretical advantage of limiting the process of dilatation and fibrosis because the thrombus is displaced while being formed, favoring the formation of a cylindrical scar from the subcutaneous and vessel wall tissues. The rope-ladder technique may have the initial advantage of favoring progressive maturation along the entire length of the fistula, but it requires fistula with sufficiently long segments suitable for cannulation. The area puncture technique weakens the fistula wall and is associated with the least favorable consequences, that is, localized dilation, disruption of the vessel wall, and subsequent development of (pseudo)aneurysms and strictures. Despite this and the fact that area cannulation has been discouraged for over two decades, it was disheartening to observe that this was the predominant practice in almost two-thirds of patients.

According to the EBPG and the Clinical Practice Guidelines for VA,

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**Figure 4 | Kaplan-Meier curves of vascular access survival according to blood flow levels, needle size, venous pressure, and cannulation technique.**
for cannulation of grafts. Specifically, according to the latter, the rotation of cannulation sites is recommended to avoid pseudoaneurysm formation. Other recommendations regarding cannulation are not available. This study showed a 22% lower risk for VA failure in those patients whose VA was cannulated with the buttonhole technique as opposed to area, confirming the results of a recently published randomized controlled clinical trial. Although the buttonhole technique is associated with good results, one should also take into consideration that it is a practice performed in centers with highly trained personnel that work with strict protocols and that it may also be used for fistulas with only short segments available for cannulation. In our study, this practice is used in 22 centers, mainly in Portugal, Turkey, the United Kingdom, and Italy. Research questions that arise from current guidelines address the effectiveness of structured cannulation training, increased remuneration for expert cannulators, and whether self-cannulation can lead to better outcomes.

Indeed, as buttonhole cannulation requires the designation of a reference nurse, especially for the initial 4–6 weeks, it is likely that this technique benefits from its association with centers offering the necessary training (i.e., centers capable of stemming the increased organizational effort and assigning the right cannulator to the right patient). In addition, once the tunnel is created, cannulation can be performed directly by patients. However, irrespective of the influence of cannulator training and center organizational issues, the underlying question to be addressed, optimally in a well-designed clinical study, is which cannulation techniques can be recommended to ensure long-term VA functionality.

This study showed that retrograde direction of arterial needle with bevel down is associated with the least favorable outcome. This is consistent with the findings of Woodson and Shapiro who reported that retrograde puncturing may be associated for an increased risk of hematoma formation, possibly owing to the related venous return of the blood (i.e., retrograde filling). Antegrade puncturing, on the other hand, may be considered fistula-protective by the same reasoning.

| Parameter                              | Category                  | Reference | HR  | 95% CI    | P-value | Marginal P-value |
|----------------------------------------|---------------------------|-----------|-----|-----------|---------|------------------|
| Age                                    | 18–50 years               | 50–65 years | 1.01| 0.86      | 1.19    | 0.91 <0.0001     |
|                                        | 65–75 years               |           | 1.03| 0.89      | 1.18    | 0.72             |
|                                        | > 75 years                |           | 1.45| 1.26      | 1.67    | <0.0001          |
| Gender                                 | Male                      | Female    | 0.93| 0.84      | 1.04    | 0.21             |
| Diabetes                               | Yes                       | No        | 1.12| 1.00      | 1.26    | 0.06             |
| Heart failure                          | Yes                       | No        | 1.39| 1.12      | 1.72    | 0.003            |
| Vintage                                | 6–24 Months               | 0–6 Months| 1.04| 0.81      | 1.33    | 0.79 0.34        |
|                                        | ≥ 24 Months               |           | 0.98| 0.77      | 1.24    | 0.84             |
|                                        | Unknown                   |           | 0.55| 0.26      | 1.18    | 0.13             |
| Platelet antiaggregation                | Yes                       | No        | 1.11| 1.00      | 1.24    | 0.05             |
| Fistula type                           | Graft                     | Fistula   | 1.74| 1.48      | 2.06    | <0.0001          |
| AV-fistula location                    | Right                     | Left      | 1.13| 1.01      | 1.27    | 0.03             |
| AV-fistula location                    | Proximal                  | Distal    | 1.49| 1.33      | 1.67    | <0.0001          |
| Needle size                            | 14 G                      | 15 G      | 1.25| 0.85      | 1.83    | 0.26 0.01        |
|                                        | 16 G                      |           | 1.21| 1.07      | 1.38    | 0.003            |
|                                        | 17 G                      |           | 1.42| 0.93      | 2.17    | 0.11             |
| Cannulation technique                  | Buttonhole                | Area      | 0.78| 0.61      | 1.00    | 0.05 0.04        |
|                                        | Rope-ladder               |           | 0.89| 0.79      | 1.00    | 0.06             |
| Bevel and needle direction             | Antegrade + bevel down    | Antegrade + bevel up | 0.97| 0.82      | 1.14    | 0.71 0.03        |
|                                        | Retrograde + bevel up     | Retrograde + bevel down | 0.93| 0.81      | 1.07    | 0.32             |
|                                        |                           |           | 1.18| 1.01      | 1.37    | 0.04             |
| Blood flow                             | < 300 ml/min              | 300–350 ml/min | 1.18| 1.01      | 1.36    | 0.03 0.03        |
|                                        | 350–400 ml/min            |           | 0.91| 0.80      | 1.04    | 0.16             |
|                                        | > 400 ml/min              |           | 0.93| 0.75      | 1.15    | 0.49             |
| Venous pressure                        | < 100 mm Hg               | 100–150 mm Hg | 1.51| 1.11      | 2.07    | 0.009 <0.0001    |
|                                        | 150–200 mm Hg             |           | 1.40| 1.20      | 1.64    | <0.0001          |
|                                        | 200–300 mm Hg             |           | 1.87| 1.54      | 2.26    | <0.0001          |
|                                        | > 300 mm Hg               |           | 2.09| 1.21      | 3.59    | 0.008            |
| Arm compression at the time of cannulation | None                  | Patient Tourniquet assistance | 1.25| 1.04      | 1.49    | 0.02 0.02        |
| Abbreviations: CI, confidence interval; HR, hazard ratio.
that is, tract closure through flow force. Therefore, retrograde direction of the arterial needle is more likely to be associated with a higher risk for aneurism. Despite recommendations by KDOQI\textsuperscript{11} to rotate the needle during insertion, the univariate analysis performed here found no evidence of any benefit of this practice. On the contrary, the authors share the opinion of many cannulators that the 180° rotation of the needle is unnecessary and may constitute an additional trauma to the VA. Further studies are needed to clarify whether rotation of the VA needle during cannulation should be recommended or not.

There are a number of possible reasons for the association of the higher failure risk with smaller needle sizes. While increased trauma and prolonged bleeding time are generally associated with the use of large needles, the use of small needles at the same blood flow results in a higher speed of the blood returning to the vasculature, possibly damaging the intima of the AVFs. For example, at an operative blood flow of 350 ml/min, the maximum speed of the injected blood will be 8.79 m/s with a 17-G needle and 5.80 m/s with a 15-G needle (presented by Ralf Jungmann at Vascular Access Course Stockholm, 11–12 October 2012, Stockholm, Sweden). Furthermore, the shear forces created by returning blood can have a role in inflammation and stenosis formation.\textsuperscript{24,25} Stenotic fistula and graft lesions are associated with the induction of the expression of profibrotic cytokines, local inflammation, and neointimal proliferation.\textsuperscript{26,27} Repeated trauma at venipuncture sites and turbulent blood flow may promote this process.\textsuperscript{25} However, we cannot exclude that this association may be a consequence of bias by indication. Needles of smaller inner dimension are generally prescribed not only for a new VA but also for problematic AVFs, that is, those likely to fail in the following months. Therefore, it is difficult to derive a conclusion from this association, but on the basis of Figure 3, 17-G needles are clearly linked to blood flow levels below 300 ml/min and, on the contrary, 14-G needles are mainly prescribed to patients with 350–400 ml/min or greater blood flows. It is also of interest to underline that higher venous pressure is mainly associated with the 16-G needles, which have a wider distribution of different blood flows. A prospective study will be required to answer these questions.

Elevated venous pressure during hemodialysis can be the first sign of access malfunction. Measurement of venous pressure during dialysis is currently used as a surveillance tool within the dialysis session, and not as a standard monitoring strategy. This study showed a significant and proportionally increasing risk of VA failure with venous pressures higher than 150 mm Hg. An increased HR was also detected for venous pressure below 100 mm Hg. As shown in Figure 3, an association between needle size, blood flow, and venous pressure is indicated in that for needle gauges 15, 16, and 17 low venous pressures appear to be associated with low blood flows. Such an association could be an indication of stenosis in the artery. Venous pressure is crucially dependent on the characteristics of the needle (e.g., the needle gauge, the length of the metallic portion, and the length and the thickness of the needle shaft), which vary among manufacturers. In this network, at the time of the study, the vast majority of the needles (85%) were from a single producer and the length of the needle was 25 mm. Consistency of the measurements can therefore be assumed. The unexpectedly high HR associated with a venous pressure of under 100 mm Hg compared with 150–200 mm Hg should motivate reflection on the currently accepted limits. One could consider integration of venous pressure monitoring into an algorithm for the detection of increased risk of access failure.

This study has certain limitations over and beyond those inherent to observational studies, for example, that residual confounding cannot be completely ruled out. Being a retrospective study, patient data for those patients on dialysis before admission to the NephroCare clinic were not collectable, and thus robust information on the number of prior VAs, on their respective lengths, and on first cannulation was not available. Particularly, the missing information on the length of the VA, its depth, and the access flow constitute a major weakness because a particular cannulation technique could have been chosen on the basis of what is possible with the given access characteristics. In addition, the length of the access can influence the way in which the needles are placed. Despite these missing data, we feel that this study has its merits, as it shows that traditional local practices have a significant influence on procedures exercised. A further limitation is that the VA practice was surveyed in April 2009 and was assumed not to have been changed during the follow-up (31 March 2012). However, as nursing practices in this field are strongly related to the clinic culture and experience, we have reason to believe that it is should not constitute a significant bias. Of course, some cannulation particulars, such as needle size and arterial blood flow, may vary over time, in that smaller needle sizes and low blood flow rate are used for initial access use and that large needles are taken for mature accesses. If this is the case, a time-varying analysis would appear indicated. However, we feel that the model selected here is also justified because it is an explanatory model, based on the association of baseline characteristics with access survival. Other limitations are that we had follow-up of 65% of the patients and that most countries were in Europe (owing to deployment of the electronic reporting system).

As reported, an association between clinical practice patterns and country has been detected, and consequently not all different practices were covered by our model. However, according to the results of this analysis, each country has a combination of practices that positively and negatively influence the VA survival. For example, in Romania, positive influences were the puncture direction being antegrade (82%), bevel orientation being predominantly upward (95%), and needle not rotated (84%); negative associations were the use of area technique (77%), preferred needle size (75% with 16 G), and the use of blood flows <300 ml/min (47%). For this reason, intracountry correlations were
considered using a sandwich estimator in the multivariate model. To assess the influence of individual center practices, we also performed a sensitivity analysis by applying the sandwich estimator at the center level. There were only negligible differences to the results obtained with the original model at the country level, raising our confidence that there is no severe confounding of the model by center practice effects.

Given the relevant impact of the investigated variables on the survival of the VA, itself a key driver of hemodialysis patient survival, we believe it is time to organize a large-scale randomized clinical trial to facilitate the formulation of practical and comprehensive cannulation practice guidelines. As the associations between practice patterns and VA survival reported here are mainly related to national procedures and only partially related to actual patient limitations, they offer some promising indications for improving clinical practice.

**MATERIALS AND METHODS**

In April 2009, a cross-sectional survey was conducted in 171 dialysis units located in Europe, the Middle East, and Africa to collect details on VA cannulation practices on a clinic by clinic level. The results have already been published. On the basis of this survey, a cohort of patients was selected for follow-up to investigate VA survival. All patients who were on double-needle hemodialysis or online hemodiafiltration during the week of the survey were selected for analysis, as long as a fistula or graft was used for VA, survey data were complete, and follow-up data were available in our clinical database.

Primary outcome was time until the first surgical access intervention resulting in the generation of a new access (i.e., as opposed to any surgical intervention done just for revision, thrombectomy, etc., or any endovascular intervention). Survey date served as baseline. Patients were censored for transplantation, death, loss of follow-up, or end of the follow-up period (31 March 2012).

Information on cannulation retrieved from the survey comprised fistula type and location, cannulation technique, needle size, needle and bevel direction, needle rotation, blood flow, arterial and venous pressure, use of disinfectants, use of local anesthesia, and application of arm compression at the time of cannulation.

To adjust for individual patient characteristics, the following information was extracted from the clinical database: patient age, gender and body mass index, prevalence of diabetes, and the use of ACE inhibitors, platelet antiaggregants, and anticoagulants. In addition, the median blood flow prescription was documented at a center level at the time of the survey.

For univariate analysis, Kaplan–Meier curves were calculated and comparisons were performed using the log-rank test. By combining univariate results with medical and statistical experience, a set of variables for multivariable analysis was determined. In particular, specific interaction terms (e.g., bevel vs. arterial needle direction) were defined for statistical examination, and decisions were made regarding their inclusion or omission in the Cox model depending on their significance or collinearity, respectively. A final Cox model based on these variables was calculated, using the sandwich estimator to account for within-country correlation. Step by step, the final model was reduced, setting a P-value of 0.1 for variable inclusion.

All analyses were performed with SAS V9.2 (SAS Institute, Cary, NC).

**DISCLOSURE**

All authors are full-time employees of Fresenius Medical Care and may hold company stock options.

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