SUMMARY

The vascular system in plants is necessary for the distribution of water and nutrients, the propagation of signalling molecules, and the mechanical support. The development of vascular system in dicot leaves is hierarchical and occurs in the continuity with the pre-existing stem vasculature. The mechanism of vasculature formation is usually explained in terms of canalization hypothesis, where the newly formed vascular strands are specified by pathways of auxin flow from auxin sources to pre-existing vascular strands (auxin sink). Thus, the aim of this study was to test experimentally a role of epidermal auxin sources and existing vasculature in the formation of the vascular pattern in growing leaf primordia of *Arabidopsis thaliana*. With using auxin-related reporters, the initiation of vascular strands has been monitoring over time in undisturbed conditions and after different (chemical, genetic, mechanical) disturbances of epidermal auxin sources and existing vasculature. The *in vivo* imaging has been coupled with clearing procedure enabling the analysis of procambium differentiation process. It has been found that the auxin source at apical primordium region does not depend on localized auxin biosynthesis, however, the PIN1-dependent polar auxin transport contributes to their maintenance. In contrast, the generation of lateral auxin sources at primordium margins is correlated to the localized auxin biosynthesis, and strongly depends on the polar auxin transport. Experimental manipulations suggest that epidermal auxin sources are not crucial for the development of the midvein in leaf primordia. Instead, it is proposed that midvein development depends on its internal auxin concentration or/and signals from connected pre-existing vasculature of the stem. Also, epidermal auxin sources are not necessary for the proper development of the vascular pattern. It is rather auxin flow or/and related auxin levels in the vascular strands that affects the overall vascular pattern in leaf primordia.