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Single chain antibodies against '*Ca*. Liberibacter asiaticus'

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Antibodies are widely used as microbiological reagents, but antibodies that recognize '*Ca*. Liberibacter asiaticus' are generally lacking. We have developed and applied immunization and affinity screening methods to create a primary library of recombinant single chain variable fragment (scFv) antibodies in an M13 vector, pKM19. The antibody population is enriched for antibodies that bind antigens of 'Ca. Liberibacter asiaticus' and *Diaphorina citri*. The primary library has more than 10^7 unique antibodies and the genes that encode them. We have screened this library of antibodies for antibodies that bind to specifically chosen proteins that are present on the surface of '*Ca*. Liberibacter asiaticus'. These proteins were used as 'bait' for affinity-based selection of scFvs that bind to the major outer membrane protein, OmpA; the polysaccharide capsule expressing protein KpsF; a protein component of the type IV pilus (CapF); and two flagellar proteins FlhA and FlgI. These scFvs have been used in ELISA and dot blot assays against purified protein antigens and '*Ca*. Liberibacter asiaticus' infected plant extracts. We also have isolated scFv that bind to surface exposed portions of the TolC proteins and of a protein called InvA. These proteins may have critical roles in pathogenicity. Thus far, screening of these scFvs is more efficient when using phage bound, rather than soluble scFvs. We have demonstrated a technology to produce antibodies and select at will and against any protein target encoded by 'Ca. Liberibacter asiaticus'. Future applications will include advanced diagnostic methods for huanglongbing and the development of immune labeling reagents for in planta applications.